

**INDUSTRY BIOSECURITY PLAN
FOR THE NURSERY & GARDEN INDUSTRY**

Threat Specific Contingency Plan

Aphid transmitted viruses

Specific examples detailed in this plan:

Plum pox potyvirus

Tobacco etch virus

Plant Health Australia

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1 Purpose and background of this contingency plan

This contingency plan provides background information on the pest biology and available control measures to assist with preparedness for an incursion into Australia of a range of viruses that are transmitted by aphids. In this contingency plan viruses have been used as examples of those considered to be of greatest economic impact and risk to the Nursery and Garden Industry. It should be noted that some aphid transmitted viruses with a high economic impact are already present in Australia.

The contingency plan provides guidelines and options for steps to be undertaken and considered when developing a Response Plan for incursion of the virus pests. Any Response Plan developed using information in whole or in part from this contingency plan must follow procedures as set out in PLANTPLAN and be endorsed by the National Management Group prior to implementation.

This contingency plan was developed for the Nursery and Industry Australia (NGIA), and therefore is focused on production nurseries covered by this association. In the event of an incursion, operations that are not covered by the NGIA or another Emergency Plant Pest Response Deed (EPPRD) signatory (e.g. retail nurseries), will not be represented or have a decision making say in any arrangements for emergency response.

The information for this plan has been primarily obtained from documents as cited in the reference section. For each virus, information on background, life cycle, host range, distribution, symptoms and management/control is given.

2 Australian nursery industry

The Australian nursery industry is a significant horticultural sector with a combined supply chain (production to retail/grower) valued at more than \$6 billion dollars annually. The industry employs approximately 45,000 people spread over more than 20,000 small to medium sized businesses including production nurseries and retail outlets. The industry is located predominantly along the Australian coastline and in major inland regions servicing urban and production horticulture.

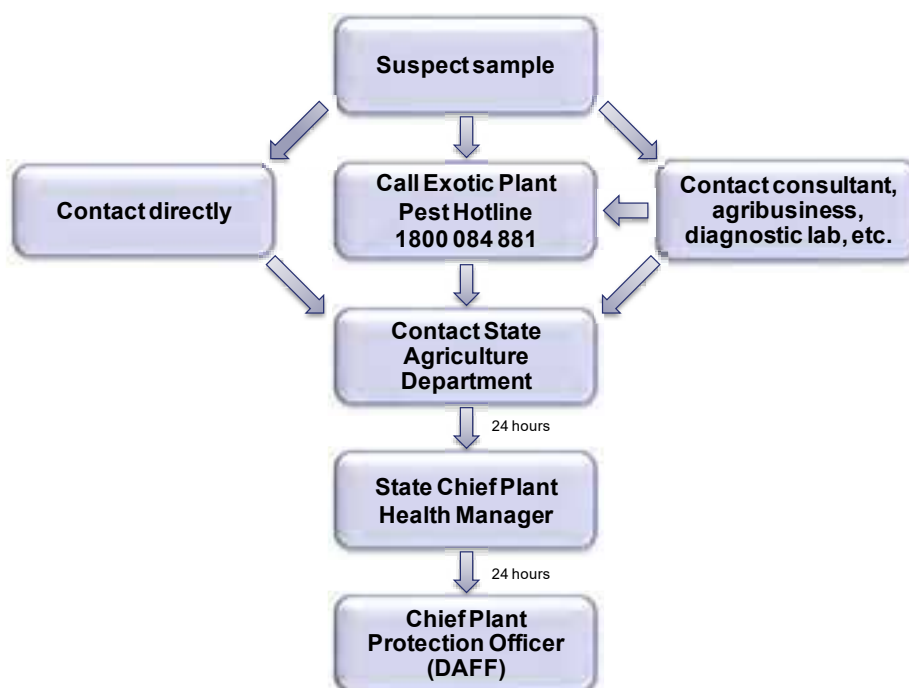
Nursery production is a highly diverse primary industry servicing the broader \$14 billion horticultural sector within Australia (Table 1). A pest incursion is likely to impact market access (see Section 11.7 Appendix 7).

Table 1. Nursery production supply sectors within Australian horticulture

Production Nursery	Horticultural markets	Economic value
Container stock ¹	Ornamental/urban horticulture	\$2 billion retail value
Foliage plants ¹	Interior-scapes	\$87 million industry
Seedling stock ²	Vegetable growers	\$3.3 billion industry
Forestry stock ³	Plantation timber	\$1.7 billion industry
Fruit and nut tree stock ²	Orchardists (citrus, mango, etc)	\$5.2 billion industry
Landscape stock ¹	Domestic & commercial projects	\$2 billion industry
Plug and tube stock ⁴	Cut flower	\$319 million industry
Revegetation stock ¹	Farmers, government, landcare	\$109 million industry
Mine revegetation	Mine site rehabilitation	Value unknown
Total horticultural market value		\$14.5 billion

2.1 Notification process for the reporting of suspect pests

Early detection and reporting may prevent or minimise the long-term impact of an incursion into Australia of aphid transmitted viruses.

**Figure 1.** Notification process for the reporting of suspect pests

¹ Data sourced from Market Monitor

² Data sourced from Horticultural Handbook 2004

³ Data sourced from ABARE 2005

⁴ Data sourced from industry

3 Eradication or containment decision matrix

The decision to eradicate should be based on the potential economic impact of host damage resulting from aphid transmitted virus infection, the cost of eradication and on technical feasibility. Eradication costs must factor in long term surveys to prove the success of the eradication program. A minimum of two years with no detections of the virus may be necessary to confirm that no aphid transmitted virus infections remain before pest free status can be declared. The timeframe needs to be considered on a case by case basis, based both on the size of the infection, the degree and distribution of the pest with the final decision determined by the National Management Group.

No specific eradication matrix has been determined for aphid transmitted viruses; however, the general decision process as outlined in Figure 2 and Table 2 should be followed in determining if an incursion of this pest will be eradicated or managed/contained. The final decision between eradication and management will be made through the National Management Group.

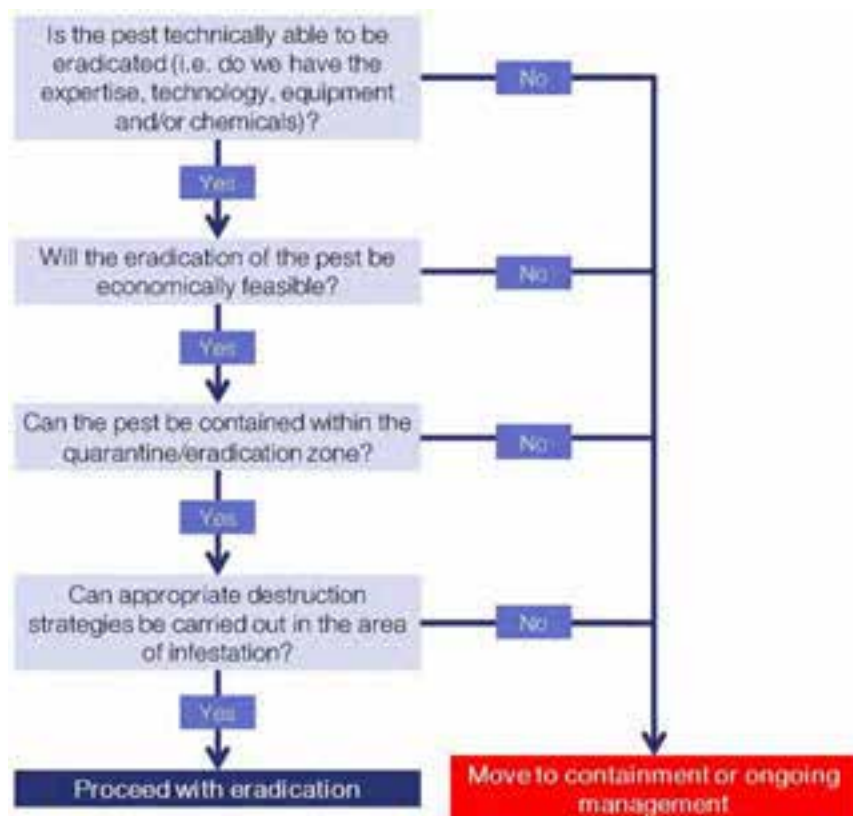


Figure 2. Decision outline for the response to an exotic pest incursion

Table 2. Factors considered in determining whether eradication or alternative action will be taken for an EPP Incident (taken from Appendix 12 of PLANTPLAN)

Factors favouring eradication	Factors favouring alternative action
<ul style="list-style-type: none"> • Cost/benefit analysis shows significant economic or amenity loss to industry or the community if the organism establishes. • Physical barriers and/or discontinuity of hosts between production districts. • Cost effective control difficult to achieve (e.g. limited availability of protectant or curative treatments). • The generation time, population dynamics and dispersal of the organism favour more restricted spread and distribution. • Pest biocontrol agents not known or recorded in Australia. • Vectors discontinuous and can be effectively controlled. • Outbreak(s) few and confined. • Trace back information indicates few opportunities for secondary spread. • Weather records show unfavourable conditions for pest development. • Ease of access to outbreak site and location of alternate hosts. 	<ul style="list-style-type: none"> • Cost/benefit analysis shows relatively low economic or environmental impact if the organism establishes. • Major areas of continuous production of host plants. • Cost effective control strategies available. • Short generation times, potential for rapid population growth and long distance dispersal lead to rapid establishment and spread. • Widespread populations of known pest biocontrol agents present in Australia. • Vectors unknown, continuous or difficult to control. • Outbreaks numerous and widely dispersed. • Trace back information indicates extensive opportunities for secondary spread. • Weather records show optimum conditions for pest development. • Terrain difficult and/or problems accessing and locating host plants.

4 Pest information/status

4.1 Pest details - viruses

A large number of viruses are spread, at least to a degree, by aphids, including viruses from the families Bromoviridae, Caulimoviridae, Comoviridae, Closteroviridae, Luteoviridae, Potyviridae, Sequiviridae and some Nanoviruses and Sobemoviruses from unassigned families (Ng and Perry 2004). Of these families, the family Potyviridae contains the majority of aphid transmitted viruses, predominantly in the genus *Potyvirus*, which is the focus of this Contingency Plan. Brault *et al.* (2010) reported that according to the International Committee of Taxonomy of Viruses the *Potyvirus* genus contains 125 definitive species and a further 88 tentative species. Many of these viruses are present and impact on crops in Australia, including *Bean common mosaic virus*, *Bean yellow mosaic virus*, *Celery mosaic virus*, *Johnson grass mosaic virus*, *Papaya ringspot virus*, *Pea seed-borne mosaic virus*, *Peanut mottle virus*, *Sugarcane mosaic virus*, *Sweet potato feathery mottle virus*, *Sweet potato virus Y*, *Turnip mosaic virus* and *Watermelon mosaic virus* (Gibbs *et al.* 2008). A list of definitive *Potyvirus* species that are exotic to Australia are listed in Table 3. Around 100 viruses tentatively classified as belonging to the genus *Potyvirus* also exist.

The *Potyvirus* group is named after the type species, *Potato virus Y*, and all viruses in this group can be transmitted by aphids (Danci *et al.* 2009). Several aphid species may transmit each *Potyvirus* and some are also transmitted in seeds (Gibbs *et al.* 2008). The viruses are transmitted mechanically by the mouthparts of aphids and are non-persistent and non-circulative (Danci *et al.* 2009). Non-circulative refers to the relatively short retention time of the virus by the vector (Brault *et al.* 2010),

meaning that the vectors only remain viruliferous for a period of minutes to hours (Ng and Perry 2004). Non-persistent transmission is characterised by short acquisition periods (seconds) and inoculation periods (minutes), where semi-persistent transmission refers to acquisition over minutes to hours, and retention of the virus by the vector for hours (Brault *et al.* 2010). For this reason, aphids can normally only carry the virus short distances; however, if strong winds are present then aphids can move large distances more quickly.

In this contingency plan specific pest information and status will be given for the aphid vector, as well as *Plum pox virus (Potyvirus)*, a virus of high risk to plum and summerfruit species, and *Tobacco etch virus (Potyvirus)*, a virus of risk to the Solanaceae family. These viruses have been chosen because of their economic importance and potential relevance to the Australian nursery industry. It should be noted however that for each virus in this family, specific information would be required on symptoms, host range, diagnosis, geographic distribution as well as risk ratings for entry, establishment, spread and economic impact.

Table 3. Exotic aphid transmitted definitive Potyviruses and their perceived importance to Australian plant industries (virus data modified from Danci *et al.* 2009).

Virus	Acronym	Industry High Priority Pest	Industry Threat Summary Tables
<i>Alstroemeria mosaic virus</i> ⁵	AIMV		
<i>Amaranthus leaf mottle virus</i>	AmLMV		
<i>Araujia mosaic virus</i>	ArjMV		
<i>Artichoke latent virus</i>	ALV		
<i>Asparagus virus 1</i>	AV-1		Nursery & Garden
<i>Azuki bean mosaic virus</i>			
<i>Bean common mosaic necrosis virus</i>	BCMNV		
<i>Beet mosaic virus</i>	BtMV		
<i>Bidens mottle virus</i>	BiMoV		
<i>Blackeye cowpea mosaic virus</i> ⁶			
<i>Calanthe mild mosaic virus</i>	CalMMV		
<i>Cardamom mosaic virus</i>	CdMV		
<i>Carnation vein mottle virus</i>	CVMoV		
<i>Carrot thin leaf virus</i>	CTLV		Vegetables
<i>Chilli veinal mottle virus</i>	ChiVMV		
<i>Cocksfoot streak virus</i>	CSV		
<i>Commelina mosaic virus</i>	ComMV		
<i>Crocus tomasinianus virus</i>			

⁵ One report in Vic in APPD

⁶ One report in Qld in APD

Virus	Acronym	Industry High Priority Pest	Industry Threat Summary Tables
<i>Cowpea green vein banding virus</i>	CGVBV		
<i>Dendrobium mosaic virus</i>			
<i>Dasheen mosaic virus</i>	DsMV		
<i>Datura shoestring virus</i>	DSTV		
<i>Dioscorea green banding virus</i>			
<i>Endive necrotic mosaic virus</i>	ENMV		
<i>Freesia mosaic virus</i>	FreMV		
<i>Garlic potyvirus</i>			
<i>Garlic virus</i>			
<i>Gloriosa stripe mosaic virus</i>	GSMV		
<i>Groundnut eyespot virus</i>	GEV		
<i>Guar green sterile virus</i>			
<i>Guinea grass mosaic virus</i>	GGMV		Grains
<i>Helenium virus Y</i>	HVY		
<i>Henbane mosaic virus</i>	HMV		
<i>Hippeastrum mosaic virus</i>	HiMV		
<i>Hyacinth mosaic virus</i>	HyaMV		
<i>Iris fulva mosaic virus</i>	IFMV		
<i>Iris mild mosaic virus</i> ⁷	IMMV		
<i>Iris severe mosaic virus</i>	ISMV		
<i>Jonquil mild mosaic virus</i>			
<i>Kalanchoe mosaic virus</i>	KMV		
<i>Konjak mosaic virus</i>	KonMV		
<i>Lily mottle virus</i>	LiMoV		
<i>Maize dwarf mosaic virus</i>	MDMV	Grains	Grains
<i>Moroccan watermelon mosaic virus</i>	WMMV		
<i>Narcissus degeneration virus</i>	NDV		
<i>Narcissus yellow stripe virus</i>	NYSV		
<i>Nerine yellow stripe virus</i>	NeYSV		

⁷ One report in Vic in APPD

Virus	Acronym	Industry High Priority Pest	Industry Threat Summary Tables
<i>Nothoscordum mosaic virus</i>	NoMV		
<i>Onion yellow dwarf</i>	OYDV		
<i>Ornithogalum mosaic virus</i>	OrMV		
<i>Parsnip mosaic virus</i>	ParMV		
<i>Pea mosaic virus</i>			
<i>Pea necrosis virus</i>			
<i>Pea seed-borne mosaic virus</i>	PSbMV		
<i>Peanut chlorotic ring mottle virus</i>			
<i>Peanut mild mottle virus</i>			
<i>Peanut stripe virus</i>			Grains
<i>Pepper mottle virus</i>	PepMoV		
<i>Pepper severe mosaic virus</i>	PeSMV		
<i>Pepper vein banding mosaic virus</i>			
<i>Pepper veinal mottle virus</i>	PVMV		
<i>Peru tomato virus</i>	PTV		
<i>Petunia flower mottle virus</i>	PFMoV		
<i>Plum pox virus</i>	PPV	Cherry, Summerfruit	Cherry, Nursery & Garden, Almonds, Summerfruit
<i>Pokeweed mosaic virus</i>	PkMV		
<i>Potato virus A</i>	PVA		Potatoes
<i>Potato virus V</i>	PVV		Potatoes
<i>Rembrandt tulip breaking virus</i>	RTBV		
<i>Sesame mosaic virus</i>	SeMV		
<i>Shallot yellow stripe virus</i>	SYSV		
<i>Sorghum mosaic virus</i>	SrMV	Sugarcane	Sugarcane, grains
<i>South African passiflora virus</i>			
<i>Statice virus Y</i>			
<i>Sweet potato A</i>			
<i>Sweet potato chlorotic leaf spot</i>			
<i>Sweet potato internal cork</i>			
<i>Sweet potato latent virus</i>	SwPLV		

Virus	Acronym	Industry High Priority Pest	Industry Threat Summary Tables
<i>Sweet potato russet crack</i>			
<i>Tamarillo mosaic virus</i>	TamMV		
<i>Telfairia mosaic virus</i>	TeMV		
<i>Tobacco etch virus</i>	TEV		
<i>Tobacco vein banding mosaic virus</i>	TVBMV		
<i>Tobacco vein mottling virus</i>	TVMV		
<i>Tulip breaking virus</i> ⁸	TBV		
<i>Tulip chlorotic blotch virus</i>			
<i>Tulip top breaking virus</i>			
<i>Vanilla necrosis virus</i>			
<i>Wakegi yellow dwarf virus</i>	WYDV		
<i>Welsh onion yellow stripe virus</i>			Onion
<i>White lupin mosaic virus</i>			
<i>Wisteria vein mosaic virus</i>	WVMV		
<i>Yam mild mosaic virus</i>	YMMV		
<i>Yam mosaic virus</i>	YMV		
<i>Zucchini yellow fleck virus</i>	ZYFV		

5 Pest information/status – aphid vectors

5.1 Pest details – aphid vectors

Taxonomic position: Kingdom, Animalia; Phylum, Arthropoda; Class, Insecta; Order, Hemiptera; Suborder, Sternorrhyncha; Family, Aphididae.

Over 200 aphid species act as vectors for plant viruses, accounting for around 50% of insect-vector-borne viruses (Nault 1997). Most aphid vectors are from the subfamily Aphidinae with a small proportion of vectors from nine other subfamilies (Ng and Perry 2004). Aphids are ideal vectors of viruses because many have wide plant host ranges, they have piercing-sucking mouthparts to facilitate the delivery of viruses into plant cells and they have high rates of reproduction, allowing disease epidemics to develop (Ng and Perry 2004).

The major aphid vectors of Plum Pox Virus (PPV) are *Aphis spiraecola* and *Myzus persicae*, and these two vector species are also two of the twelve known vectors of Tobacco etch virus (TEV). Thus,

⁸ Two records of this in Vic in APPD, one was at Knoxfield.

this contingency plan will focus on these two vectors (both of which are present in Australia) as examples of aphid vectors of viruses (Table 4).

Table 4. Examples of aphid vectors of exotic virus threats to the Nursery & Garden Industry

Scientific name	Synonyms	Common names
<i>Aphis spiraecola</i>	<i>Anuraphis erratica</i> del Guercio, 1917, <i>Aphis bidentis</i> Theobald, 1929, <i>Aphis citricola</i> van der Goot, 1912, <i>Aphis croomiae</i> Shinji, 1922, <i>Aphis deutziae</i> Shinji, 1922, <i>Aphis malvoides</i> van der Goot, 1917, <i>Aphis mitsubae</i> Shinji, 1922, <i>Aphis nigricauda</i> van der Goot, 1917, <i>Aphis pirifoliae</i> Shinji, 1922, <i>Aphis pseudopomi</i> Blanchard, 1939, <i>Aphis pseudopomi</i> Bertels, 1973, <i>Aphis virburnicolens</i> Swain, 1919	Green citrus aphid Spirea aphid
<i>Myzus persicae</i>	<i>Aphis convolvuli</i> Kaltenbach, 1843, <i>Aphis cynoglossi</i> Walker, 1848, <i>Aphis derelicta</i> Walker, 1849, <i>Aphis dianthi</i> Schrank, 1801, <i>Aphis dubia</i> Curtis, 1842, <i>Aphis egressa</i> Walker, 1849, <i>Aphis malvae</i> Mosl., 1841, <i>Aphis persicae</i> Sulzer, 1776, <i>Aphis persiciphila</i> , <i>Aphis persola</i> Walker, 1848, <i>Aphis rapae</i> Curtis, 1842, <i>Aphis redundans</i> Walker, 1849 sec. Laing, 1925, <i>Aphis suffragans</i> Walker, 1848, <i>Aphis tuberoscellae</i> , <i>Aphis vastator</i> , <i>Aphis vulgaris</i> Kyber, 1815 (sec. Walker), <i>Aulacorthum convolvuli</i> , <i>Myzodes persicae</i> (Sulzer), <i>Myzodes tabaci</i> Mordvilko, 1914, <i>Myzoides persicae</i> , <i>Myzus dianthi</i> (Schrank), <i>Myzus malvae</i> Oestl., 1886 (sec. Theob.), <i>Myzus nicotianae</i> Blackman, <i>Myzus pergandei</i> Sanders, 1901 sec. Patch, <i>Myzus persicae</i> var. <i>cerastii</i> Theobald, <i>Myzus persicae</i> var. <i>sanguisorbella</i> Theobald, 1926, <i>Nectarosiphon persicae</i> (Sulzer), <i>Phorodon cynoglossi</i> Williams, 1891 sec. Davis, 1911, <i>Phorodon persicae</i> (Sulzer), <i>Rhopalosiphum betae</i> Theobald, 1913, <i>Rhopalosiphum calthae</i> Koch, 1854, <i>Rhopalosiphum dianthi</i> , <i>Rhopalosiphum lactucellum</i> , <i>Rhopalosiphum lactucellum</i> Theobald, 1915, <i>Rhopalosiphum persicae</i> , <i>Rhopalosiphum solani</i> Theobald, 1912 nonKalt, 1843, <i>Rhopalosiphum tuberosellae</i> Theobald, 1922, <i>Rhopalosiphum tulipae</i> Thos., 1879 sec. Davis, 1911, <i>Siphonophora achyranthes</i> Mon., 1879, <i>Siphonophora nasturtii</i> Koch, 1855	Green peach aphid

5.1.1 Background

Both *Myzus persicae* and *Aphis spiraecola* are widespread in Australia and are pests of many agriculturally important crops. They cause damage to plants directly as their piercing-sucking mouthparts are inserted into the plant tissue to feed on leaves, green shoots and flowers. Leaves may curl as a result of feeding damage, and the large quantities of honeydew produced may result in blackening of leaves and fruit due to sooty mould.

5.1.2 Life cycle

Aphids can propagate parthenogenically, that is, females do not need to mate to produce young and no eggs are laid. Sexual and parthenogenic reproduction alternate in the life cycle, with sexual forms typically appearing in autumn to oviposit overwintering eggs on the primary host (Komazaki 1993). Eggs hatch in spring, and each hatched larva develops into a mother which reproduces parthenogenically (Komazaki 1993). Adult females can be wingless or winged, with the presence of wings indicating a decline in food quality or overcrowding (Broughton 2007).

Development of *M. persicae* can be rapid, often 10 to 12 days for a complete generation though in cooler temperatures the life cycle may last up to 50 days (Toba 1964). Fertility drops rapidly at temperatures over 30°C, but on average each female gives rise to 50-80 nymphs (wingless forms lay more eggs than winged forms). Over 20 annual generations have been reported in mild climates.

Further, longevity of *M. persicae* may be affected by temperature, type of life cycle (egg laying or live births), and plant host (Mau and Kessing 1991). In warmer climates, reproduction does not involve mating and egg laying i.e. the entire life cycle is parthenogenic (Mau and Kessing 1991). As a consequence of this type of reproduction, populations are composed solely of females. In colder climates where suitable host plants cannot persist, aphids overwinter in the egg stage on *Prunus* spp.

For *A. spiraecola*, an entire generation can develop within a week and there can be up to 25 generations per year, with females producing 60 young each (Broughton 2007). Up to 40 generations per year have been reported in Italy. Wang and Tsai (2000) reported that the mean generation time of *A. spiraecola* ranged from 35.1 days at 10°C to 10.7 days at 32°C and the optimal range of temperature for population growth was 20-30°C.

Where sexual reproduction is apparent in *A. spiraecola*, the primary hosts are *Spiraea* or *Citrus*. However, over the majority of its geographical range its reproduction is entirely parthenogenic (CABI 2011).

5.1.3 Dispersal

Both *A. spiraecola* and *M. persicae* can be transported long distances by wind and storms, and as such are found throughout the world and in all parts of Australia.

5.2 Affected hosts

5.2.1 Host range

M. persicae is polyphagous with host plants in over 40 families. *A. spiraecola* also has a wide host range across multiple families. Lists compiled in the cabicompendum (CABI 2011) for hosts of *M. persicae* and *A. spiraecola* are presented in Sections 11.1 and 11.2, respectively.

5.2.2 Current geographic distribution

Both *M. persicae* and *A. spiraecola* have a worldwide distribution, including having a widespread distribution in Australia.

5.2.3 Symptoms

Both species have toxic saliva and can cause direct damage to plants. Symptoms can include dwarfing, wilting or curling of leaves, while high aphid densities can lead to water stress, reduced growth and ultimately to decreased yield.

5.3 Diagnostic information

5.3.1 *M. persicae*

The wingless adult aphids vary in colour from green to pale yellow, or pink, red or black, and are 1.5 to 2 mm long (Mau and Kessing 1991). Winged adults have green abdomens with black or dark brown markings, a black thorax and translucent wings (Figure 3).

Nymphs are pale yellowish-green in colour with three dark lines on the back of the abdomen. Nymphal development is completed in 6 to 11 days in warmer climates (Toba 1964). In cooler regions, aphids overwinter during the egg stage. The eggs are shiny black and are often laid on the bark of fruit trees.



Figure 3. *Myzus persicae* winged adult. Image courtesy Scott Bauer, USDA Agricultural Research Service, Bugwood.org.

5.3.2 *A. spiraecola*

Adults are apple green with brown heads, mainly pale legs and antennae, 2 mm in length and can be winged or wingless. Nymphs are pear shaped, apple green to bright yellow in colour (Broughton 2007) (Figure 4).



Figure 4. *Aphis spiraecola*. Image courtesy Andrew Jensen

5.4 Pathogen risk ratings and potential impacts

Both *M. persicae* and *A. spiraecola* are widespread within Australia, and cause damage to crops directly as well as act as vectors for the spread of viruses.

6 Pest information/status – aphid transmitted viruses

6.1 Pest details – example: Plum pox virus

Common name:	Plum pox virus (PPV)
Scientific name:	<i>Plum pox virus (Potyvirus)</i>
Synonyms:	Peach sharka, Pox disease of plum, Sharka, Sharka disease of plum
Taxonomic position:	Family, Potyviridae; Genus, Potyvirus

The information from this plan has been primarily obtained from documents as cited in the reference section as well as material sourced from the 'Diagnostic Protocol for *Plum pox virus* (PPV)' (SPHDS, 2010).

6.1.1 Background

Plum pox virus (PPV) has been chosen as an example in this contingency plan as it is one of the most destructive diseases of stone fruits and has a very wide host range among *Prunus* species. It is

not present in Australia but has been introduced to many other countries from its origin in eastern Europe (Bulgaria). It has been eradicated, or is currently under eradication or containment, from several countries.

PPV is a filamentous virus with particles 750 nm long and 15 nm in diameter. The virus has a single-stranded RNA genome with a molecular weight of 3.5×10^6 Da. Protein inclusions of the pinwheel type are present in the cytoplasm of infected host cells (Salvador *et al.* 2006).

Currently, PPV can be divided into six strains: PPV-D, PPV-M, PPV-EI Amar, PPV-C, PPV-W, and PPV-Rec (Candresse and Cambra 2006; James and Glasa 2006). Strains of PPV were originally distinguished on the basis of symptoms induced in herbaceous indicator plants. Kerlan and Dunez (1979) then serologically differentiated Dideron (PPV-D) and Markus (PPV-M) strains, the former on apricot (*Prunus armeniaca*) in France and the latter originally on peach (*Prunus persica*) in Greece. Within these strains, individual isolates can vary in the severity of symptoms they induce.

PPV-D is the most common strain in Western Europe and is the only strain currently found in the US and Canada (Levy *et al.* 2000).

PPV-M is considered to be a more aggressive, epidemic form and is the most common strain in Southern, Eastern and Central Europe. Peach is most susceptible to PPV-M. An isolate of the PPV-M strain which is very aggressive and necrogenic on peach was reported in France in the 1980s (Candresse *et al.* 1993). This necrogenic strain has since been referred to as PPV-SP.

The EI Amar strain (PPV-EI Amar) is distinct from the D and M strains on the basis of divergences in RNA sequence (Wetzel *et al.* 1991a) and has only been isolated in the North Africa region.

The cherry strain (PPV-C) has been found to infect sweet (*Prunus avium*) and sour (*Prunus cerasus*) cherry trees and is found in Eastern and Central Europe (Kalashyan *et al.* 1994; Kölber *et al.* 1997). It differs significantly from other strains of PPV in biological, serological and molecular properties.

PPV-Rec is a stable recombinant strain consisting of D and M strain recombinants with a common phylogenetic link (Glasa *et al.* 2004). It has been reported in several European countries, often having been incorrectly identified as PPV-M.

A sixth strain of PPV, termed PPV-W has been identified in Canada (James *et al.* 2003; James and Varga 2005).

6.1.2 Life cycle

Aphid vectors probe or feed on infected trees, then fly to other trees where they again probe or feed. The aphids can acquire the virus from infected leaves, flowers, or fruits in seconds to minutes and can transmit it to new plants within, again, only a few minutes. There is no latent period in the insect.

After inoculation, an incubation period occurs and infected plants may not show symptoms for several months. Symptoms are often transient and may easily be mistaken for those of other diseases allowing the virus to become established before the disease is recognised as PPV. Systemic spread of the virus within woody hosts may take several years (OEPP/EPPO 1983). The virus may be distributed very irregularly throughout the tree further confounding detection.

Infection begins to spread from the first infected trees after 2-3 years (Llácer *et al.* 1986).

6.1.3 Dispersal

PPV is transmitted in a non-persistent manner by two main winged aphid vectors, *Aphis spiraecola* and *Myzus persicae*, both of which are widespread throughout Australia. Other aphids that can transmit PPV, but less efficiently, include *A. craccivora*, *A. fabae*, *A. gossypii*, *A. hederiae*, *Brachycaudus cardui*, *B. helvichrysi*, *B. persicae*, *Hyalopterus pruni*, *M. varians*, *Phorodon humuli*, and *Rhopalosiphum padi* (Avinent *et al.* 1994; Kunze and Krczal 1971; Labonne *et al.* 1994; Leclant 1973). *Metopolophium dirhodum* and *Toxoptera citricida* have been shown to be vectors of PPV under experimental conditions (Gildow *et al.* 2004).

The number of trees becoming infected in an orchard is directly related, in a given season, to numbers of these aphid vectors. Aphids spread the disease to trees several spaces away rather than to immediately adjacent trees (Gottwald *et al.* 1995).

Grafting can also contribute significantly to the spread of PPV in infected areas if certified virus-free material is not used. Dissemination of the virus nationally and internationally is most often through transport of uncertified plants or plant parts (Diekmann and Putter 1996).

Although spread is difficult to control within a localised area as aphid vectors have been shown to carry the virus for several kilometres if starved (Levy *et al.* 2000). Long distance spread can be controlled through the use of certified virus-free nursery stock and strict quarantine regulations.

6.1.4 Host range

The major hosts of PPV are fruit-producing species of the *Prunus* genus and Levy *et al.* 2000 reports the following as having been shown to be natural hosts:

- *P. armeniaca* - Apricot
- *P. persica* - Peach
- *P. persica* var. *nectarina* - Nectarine
- *P. domestica* - Garden plum (prune)
- *P. salicina* - Japanese plum
- *P. insititia* - Damson plum
- *P. cerasifera* - Myrobalan plum
- *P. glandulosa* - Dwarf flowering almond, Cherry almond
- *P. avium* - Sweet cherry
- *P. cerasus* - Sour (tart) cherry
- *P. dulcis* - Almond

While almond (*P. dulcis*) can be infected with PPV, it expresses few, if any, symptoms (Llácer and Cambra 2006).

It is also thought that many other cultivated or weedy annual plant species can potentially carry PPV and Section 11.3 Appendix 3 provides a list of PPV's host species (CABI 2011). An asterisk (*) indicates those species that are often grown in domestic gardens or used as street trees in Australia. These species would need to be considered in the event of an incursion of PPV (Diagnostic protocol for *Plum pox virus*).

6.1.5 Current geographic distribution

PPV is widespread throughout Europe and is found in Albania (restricted distribution), Austria (restricted distribution), Bosnia-Herzegovina, Bulgaria, Croatia, Cyprus (restricted distribution), the Czech Republic, France (restricted distribution), Germany, Greece, Hungary, Italy (restricted distribution), Latvia (under eradication), Lithuania, Luxembourg, Moldova (restricted distribution), Montenegro, the Netherlands, Norway (restricted distribution), Poland, Portugal (restricted distribution), Romania, Russia (restricted distribution), Serbia, Slovakia, Slovenia (restricted distribution), Spain (restricted distribution), Switzerland, Syria, Turkey (restricted distribution), Ukraine (restricted distribution), and the United Kingdom (restricted distribution) (CABI 2007). PPV is also present in Argentina, Canada (restricted distribution), Chile, China, Egypt, India (restricted distribution), Iran, Jordan, Kazakhstan, Pakistan, Tunisia, the United States of America (restricted distribution) (CABI 2011).

The virus was present in but has since been eradicated from Belgium, Denmark, Estonia, and Georgia (CABI 2011).

6.1.6 Symptoms

Symptoms of PPV depend very much on the strain of the virus; the host species, cultivar and plant tissue infected; as well as locality and season (Dosba *et al.* 1986; Kegler and Hartmann 1998).

Symptoms may appear on leaves, flowers, fruit, or seeds. Diseased apricots and plums are deformed with a misshapen, bumpy appearance. Apricots show internal browning of the flesh with stones displaying pale spots or rings. Infected fruit may drop prematurely (Dunez 1987) (Figure 5).

Symptoms are particularly conspicuous on leaves of all *Prunus* in spring and include vein clearing or chlorotic bands, spots, or rings (Figure 6). Infected fruit show spots or rings that can be lighter or darker in colour than the surrounding skin.

In peaches, leaf deformation resembling insect damage can sometimes be observed. An early symptom of PPV infection in peaches is the appearance of dark pink streaks on flower petals and this is the most reliable symptom to look for in this host. Necrotic areas can also be seen on peach leaves.

Cherry leaves show mottling and necrotic spots.

In the case of plums, leaf spots or rings can be large or small and speckle-like with some necrotic areas on leaves that often fall out giving a shot-hole appearance. The fruit can show large or small and speckle-like spots or rings on the surrounding skin that may also develop sunken lesions (Figure 7).



Figure 5. Symptoms of PPV infection on apricots (*P. armeniaca*) fruit and stones. Image courtesy of Ministry of Agriculture and Regional Development Archive, Ministry of Agriculture and Regional Development, Bugwood.org.



Figure 6. Symptoms of PPV infection on apricot (*P. armeniaca*) leaves. Image courtesy of Biologische Bundesanstalt für Land- und Forstwirtschaft Archive, Biologische Bundesanstalt für Land- und Forstwirtschaft, Bugwood.org



Figure 7. Symptoms of PPV infection on plum (*P. domestica*) fruit. Image courtesy of Biologische Bundesanstalt für Land- und Forstwirtschaft Archive, Biologische Bundesanstalt für Land- und Forstwirtschaft, Bugwood.org.

6.1.7 Diagnostic information

An endorsed National Diagnostic Protocol (NDP2) for Plum pox virus (PPV) has been prepared (SPHDS 2010). This protocol describes three methods for the positive identification of PPV including electron microscopy, ELISA, and RT-PCR and a short summary of each method is outlined below. For further information on standard diagnostic protocols see Section 11.4 Appendix 4. For a full list of diagnostic facilities and advisory services that can be utilised in the event of an incursion see Section 11.5 Appendix 5.

6.1.7.1 ELECTRON MICROSCOPY

Transmission electron microscope (TEM) examination of grids prepared from small sections of homogenised leaf tissue negatively stained with 1% uranyl acetate (UA) can be used to rapidly detect PPV. The grids are examined at 40,000 x magnification for flexuous filamentous *Potyvirus* particles 660-770nm in length. Immunosorbent electron microscopy (ISEM) can be used when the concentration of virus particles in the crude sap preparation is low. Two detailed protocols for crude sap preparation, one of which being specific to ISEM, are described in the National Diagnostic Protocol for *Plum pox virus* (PPV).

Diagnosis using electron microscopy requires validation by ELISA and/or RT-PCR.

6.1.7.2 ELISA

The National Diagnostic Protocol for *Plum pox virus* (PPV) describes a protocol for double antibody sandwich enzyme linked immunosorbent assay (DAS-ELISA) which enables the serological detection and quantification of PPV capsid protein using both monoclonal and polyclonal antibodies. The

antibodies selected for use allow for the detection of PPV-D, PPV-M, PPV-EI Amar, PPV-C, and PPV-W strains of the virus.

Due to its specificity and high throughput, DAS-ELISA can be used to specifically target PPV during surveys in production areas, commercial nurseries, or for production purposes.

6.1.7.3 RT-PCR

The reverse transcription polymerase chain reaction (RT-PCR) is a rapid, specific, and sensitive test that can be used to detect and diagnose PPV from extracted nucleic acids. The National Diagnostic Protocol for *Plum pox virus* (PPV) describes a two-step RT-PCR reaction which allows the cDNA template obtained to be stored for future RT-PCR testing and subsequent cloning. Standard primers designed for the detection of all strains of PPV (Wetzel *et al.* 1991b) are used. A restriction digestion test can then be used to differentiate between the strains.

While more expensive than ELISA and less suitable as a high throughput method, RT-PCR is very specific and sensitive and allows for the detection of minimal amounts of target RNA. As such, RT-PCR can be used to validate the results from electron microscopy and ELISA. These results can be further validated by sequencing the DNA amplicon and using the Basic Local Alignment Search Tool (BLAST) to align the sequence obtained with sequences held in the NCBI-GenBank database.

6.1.7.4 INDICATOR SPECIES FOR PPV

The following are important herbaceous indicator or propagation hosts of Plum pox virus (taken from Levy *et al.* 2000):

- *Chenopodium foetidum*
- *Nicotiana benthamina*
- *N. bigelowii*
- *N. clevelandii*
- *N. occidentalis* #37 B
- *N. edwardsonii*
- *N. megalosiphon*
- *N. tabacum*
- *N. physalodes*
- *Pisum sativum* cv. Colmo

6.1.8 Risk assessments for pathways and potential impacts – Plum pox virus

6.1.8.1 ENTRY POTENTIAL

Rating: Low

Australia has been importing apricots, peaches and nectarines, plums, and cherries from New Zealand since 1986 and cherries from the USA since 1996, and in 2010 Biosecurity Australia gave approval for the import of peaches and nectarines and plums. As PPV is not present in New Zealand or in the areas of production in the USA (California, Idaho, Oregon, and Washington), it is unlikely that PPV would be introduced to Australia via these pathways. While Chile, China, Japan, Mexico, and South Africa have requested market access and PPV is present in both Chile and China, it should be noted that introduction of PPV on fruit is considered to be very low.

The most likely pathway by which PPV could be introduced to Australia is through the importation of infected but symptomless budwood for grafting. The risk of entry of PPV into Australia is therefore **Low**.

6.1.8.2 ESTABLISHMENT POTENTIAL

Rating: High

Due to the fact that symptoms do not appear for several months, establishment of PPV can occur prior to detection. The virus multiplies in growing host plants. Almond, apricot, nectarine, peach and plum are common trees in Australia and aphid species are present in Australia that can spread PPV. As such, the likelihood of PPV establishing is considered to be **High**.

6.1.8.3 SPREAD POTENTIAL

Rating: High

Due to the wide host range available in Australia, the presence of aphid vectors, and lack of a latent period within the aphid vectors, the potential for spread of PPV following establishment is **High**. Aphids have been shown to carry the virus for several kilometres if starved (Levy *et al.* 2000). In addition, the climate, vector and host numbers in temperate areas of Australia would be similar to those in areas where the virus is prevalent and would therefore be suitable for the spread of PPV.

6.1.8.4 ECONOMIC IMPACT

Rating: High

PPV can significantly reduce fruit yield and quality, rendering fruit unpalatable and unmarketable. As such, the virus has a major economic impact on stonefruit production and international trading worldwide. The global cost associated with the management of PPV worldwide, excluding indirect trade costs, has been estimated to be 10 billion euros (Cambra *et al.* 2006). The economic impact of PPV in Australia could be **High**.

6.1.8.5 ENVIRONMENTAL IMPACT

Rating: Negligible

Infected fruit does not pose a health problem for humans or animals, and native plants are not known to be hosts of PPV so the environmental impact is considered to be **Negligible**.

6.1.8.6 OVERALL RISK

Rating: Medium

Based on the individual ratings above, the combined overall risk is considered to be **Medium**.

6.2 Pest details – example: Tobacco etch virus

Common name:	Tobacco etch virus (TEV)
Scientific name:	<i>Tobacco etch virus (Potyvirus)</i>
Taxonomic position:	Family, Potyviridae; Genus, Potyvirus

6.2.1 Background

Tobacco etch virus (TEV) is a disease of Solanaceous species including tomatoes and capsicums. TEV has been chosen as an example virus in this contingency plan as these Solanaceous species make up a significant component of seedling production in nurseries and this virus would be expected to have a high economic impact on these crops should it enter Australia.

TEV is a flexuous filamentous virus with particles 730-750 nm long and 12-13 nm in diameter and a single-stranded RNA genome (Purcifull and Hiebert 1982). It induces the formation of crystalline nuclear inclusions and cytoplasmic, cylindrical (pinwheel) inclusions in host cells.

The virus is transmitted in a non-persistent manner by the aphid vector, *Myzus persicae*, which is widespread throughout Australia. It can also be transmitted by several other aphid species (AVRDC 2004; 2005). The virus is also readily transmitted mechanically and by grafting using infected plant material.

6.2.2 Life cycle

Aphid vectors feed on infected plants then fly to other plants where they again probe or feed. The aphids can acquire the virus from feeding on an infected plant for less than a minute and can transmit it to a new plant just as quickly (AVRDC 2005). The aphids will retain the virus for a day or longer if the aphid does not feed after acquisition of the virus.

TEV overwinters in perennial Solanaceous weeds and is transmitted to tobacco, tomato, and pepper plants by migrating aphids.

TEV often occurs in combination with other aphid-borne viruses.

6.2.3 Dispersal

TEV is not reported to be seedborne in any plant species (Purcifull and Hiebert 1982). Its principal mode of transmission is by *M. persicae* and several other aphid species in a non-persistent manner (AVRDC 2004; 2005). The virus is also readily transmitted mechanically and by grafting.

Although spread is difficult to control within a localised area due to aphid vectors, long distance spread can be controlled through the use of certified virus-free nursery stock and strict quarantine regulations.

6.2.4 Host range

The major hosts of TEV are species of the Solanaceae family and include Tobacco (*Nicotiana tabacum*), Tomato (*Solanum lycopersicum*), and Bell and Tabasco peppers (*Capsicum annuum*, *C. frutescens*). Various weeds are also affected (AVRDC 2004; 2005). A detailed list of host plants is given in Table 5.

Table 5. Hosts of TEV as listed on the CAB Compendium (CAB International 2011)

Scientific name	Common name
<i>Capsicum annuum</i>	Bell pepper
<i>Capsicum frutescens</i>	Tabasco pepper
<i>Chenopodium album</i>	Fat hen
<i>Cirsium vulgare</i>	Spear thistle
<i>Datura stramonium</i>	Jimson weed
<i>Linaria canadensis</i>	Blue toadflax
<i>Nicotiana tabacum</i>	Tobacco
<i>Physalis heterophylla</i>	Clammy ground cherry
<i>Senna obtusifolia</i>	Sicklepod
<i>Solanum carolinense</i>	Carolina horsenettle
<i>Solanum lycopersicum</i>	Tomato
<i>Solanum seaforthianum</i>	Star potato-vine
<i>Solanum viarum</i>	Tropical soda apple

6.2.5 Current geographic distribution

TEV is found primarily in the Western Hemisphere, especially in North and South America (CABI 2011). The virus has been reported in Canada, Mexico, Puerto Rico, the USA including Hawaii, and Venezuela (CABI 2011).

6.2.6 Symptoms

The severity of symptoms in cultivated plants depends greatly on the host species and cultivar, virus strain, and time of infection (AVRDC 2004).

In tobacco, affected plants are stunted. The leaves are narrowed and show vein-clearing, mottling and a characteristic etching pattern which may become necrotic (Johnson 1930; Shepherd and Purcifull 1971) (Figure 8). Symptoms and effects on yield can be more severe in Burley (air-cured) tobaccos than flue-cured types (Stover 1951).

All stages of growth are affected in infected pepper plants. Symptoms on leaves include vein-clearing and vein-banding, mosaic patterns which fade to mottling as the leaf ages, and narrowing and distortion of leaves (Johnson 1930; McLean 1962; Zitter 1971, Figure 9 and Figure 10). Plants infected early have shortened internodes and can be severely stunted (AVRDC 2004). Fruit from such plants is small, misshapen, and displays severe mosaic symptoms. TEV can cause root necrosis, wilting, and death in Tabasco pepper plants (Greenleaf, 1953). The symptoms of TEV on pepper plants may be confused with those of other viruses such as Pepper mottle virus (PMV) or Potato virus Y (PVY) (AVRDC 2004). It should be noted that cultivars of pepper species with resistance to TEV have been developed (Cook *et al.* 1976).

As with pepper, all stages of growth in infected tomato plants may be affected. Leaves may show mild vein-clearing and mottling, a reduction in size, and distortion such as crinkling and pronounced downward curling (McLean, 1962; AVRDC 2005). Plants infected early have shortened internodes and can be severely stunted (Debrot 1976; Zitter and Tsai 1981). Fruit from such plants is mottled and does not reach full size (Figure 11). The symptoms of TEV on tomato plants may be confused with those of other viruses such as Tomato mosaic virus (ToMV) or Potato virus Y (PVY) (AVRDC 2005). However, due to the fact that several varieties of tomato carry partial resistance to ToMV while none have been bred for resistance to TEV or PVY, mottling tends to indicate infection with either TEV or PVY. Symptoms of TEV are usually more severe than those of PVY which usually consist of only faint mottling and slight distortion of leaves.



Figure 8. Symptoms of TEV on tobacco (*N. tabacum*). Image courtesy of R.J. Reynolds Tobacco Company Slide Set, R.J. Reynolds Tobacco Company, Bugwood.org.



Figure 9. Symptoms of TEV on bell pepper (*C. annuum*). Image courtesy of Florida Division of Plant Industry Archive, Florida Department of Agriculture and Consume Services, Bugwood.org.



Figure 10. Symptoms on *Capsicum annuum* leaves showing dark green mosaic areas associated with veins. Image courtesy of Thomas A. Zitter, Cornell University.



Figure 11. Symptoms on tomato showing mottling fruit caused by infection with TEV. Courtesy of Thomas A. Zitter, Cornell University.

6.2.7 Diagnostic information

No National Diagnostic Protocol has been developed or endorsed for TEV.

6.2.7.1 ELISA

Agdia markets a double antibody sandwich enzyme linked immunosorbent assay (DAS-ELISA) kit which enables the serological detection and quantification of TEV using polyclonal antibodies. A detailed protocol is available from the Agdia website (<http://www.agdia.com/>).

6.2.7.2 RT-PCR

The reverse transcription polymerase chain reaction (RT-PCR) is a rapid, specific, and sensitive test that can be used to detect and diagnose TEV from extracted nucleic acids. Carrasco *et al.* (2007) describes a set of primers used in real-time RT-PCR assays to quantify the fitness of TEV. Alternatively, Sangrok Biolab markets an immunocapture RT-PCR kit which includes pre-coated PCR tubes for capturing virus particles which can then be directly amplified by RT-PCR. A detailed protocol is available from the Sangrok Biolab website (<http://www.srbiolab.com/>).

RT-PCR can be used to validate the results from ELISA. These results can be further validated by sequencing the DNA amplicon and using the Basic Local Alignment Search Tool (BLAST) to align the sequence obtained with sequences held in the NCBI-GenBank database.

6.2.8 Risk assessments for pathways and potential impacts - TEV

6.2.8.1 ENTRY POTENTIAL

Rating: Low

Australia imports tomato and bell pepper seeds but, as TEV is not seed borne, it is unlikely that TEV would be introduced to Australia via this pathway. It is more likely that TEV could be introduced via an aphid vector entering the country with a cold-stored shipment of an alternate host of the vector. The risk of entry of TEV into Australia is considered to be **Low**.

6.2.8.2 ESTABLISHMENT POTENTIAL

Rating: High

TEV host plants and vectors are widespread in Australia, and the ability of the virus to become established is expected to be **High**.

6.2.8.3 SPREAD POTENTIAL

Rating: High

Due to the wide host range available in Australia including weeds, the presence of aphid vectors, and the fact that the virus is also transmitted mechanically, the potential for spread of TEV following establishment is considered to be **High**.

6.2.8.4 ECONOMIC IMPACT

Rating: High

TEV is responsible for heavy losses in both tomato (Zitter and Tsai 1981) and pepper (Zitter and Ozaki 1973) crops. A 25% reduction in yield has been reported for tomatoes in Venezuela (Debrot 1976). The economic impact of TEV in Australia in tobacco, tomato and capsicum and through nursery production that supplies seedlings to these industries is expected to be **High**.

6.2.8.5 ENVIRONMENTAL IMPACT

Rating: Negligible

There is no known potential to degrade the environment or otherwise alter the ecosystem by affecting species composition or reducing the longevity or competitiveness of wild hosts.

6.2.8.6 OVERALL RISK

Rating: Medium

Based on the individual ratings above, the combined overall risk is considered to be **Medium**.

7 Pest management

7.1 Response checklist

The following checklist (Table 6) provides a summary of generic requirements to be identified and implemented within a Response Plan for an incursion of a new aphid transmitted virus into Australia.

Table 6. Checklist of requirements to be identified in a Response Plan

Checklist item	Further information
Destruction methods for plant material, soil and disposable items	Section 8.1.1, 8.1.2
Disposal procedures	Section 8.1.5
Quarantine restrictions and movement controls	Section 8.3
Decontamination and property cleanup procedures	Section 8.5
Diagnostic protocols and laboratories	Section 5.3
Trace back and trace forward procedures	Section 8.6
Protocols for delimiting, intensive and ongoing surveillance	Section 7.2
Zoning	Section 8.4
Reporting and communication strategy	Section 11.6

A range of specifically designed procedures for the emergency response to a pest incursion and a general communication strategy refer to PLANTPLAN (Plant Health Australia 2010). Additional information is provided by Merriman and McKirdy (2005)⁹ in the Technical Guidelines for Development of Pest Specific Response Plans.

7.2 Surveys and epidemiology studies

Information provided in Sections 7.2.1 to 7.2.3 provides a framework for the development of early detection and delimiting surveys for aphid transmitted viruses.

Where aphids are found in a production nursery that is in close proximity to potential host plants, periodically inspect nearby hosts for signs of virus symptoms by examining stems, leaves and fruit. Some hosts may be asymptomatic and leaf samples may be required from all known hosts as part of a targeted survey.

Infested sources within the production nursery may provide an opportunity for aphids to spread to trees and shrubs outside the production nursery. Personnel should avoid moving plant material between production nurseries to limit movement of both the vector and virus infected material. Shoes, tools and vehicle tyres should be thoroughly washed of soil and then sanitised with a registered disinfectant. Extra precaution should be taken when working in areas known to be infested, including disposable overboots that may be used and disposed of on-site.

⁹ Available on the PHA website (www.planthealthaustralia.com.au/go/phau/biosecurity/general-biosecurity-information)

7.2.1 Technical information for planning surveys

When developing surveys for presence and/or distribution of the virus (and potentially for the virus vectors), the following characteristics provide the basic biological knowledge that informs the survey strategy:

- Virus infected plant material may be asymptomatic
- Host species in Australia are likely to be numerous and widely dispersed and may be present within relevant industries, nurseries as well as home gardens, landscape plantings and weeds
- Numerous aphid vectors are already present and widespread in Australia and many aphids (including *M. persicae* and *A. spiraecola*) have wide host ranges and share many of the same hosts with that of the virus
- The risk of aphid movement on nursery stock machinery, equipment and personal effects is high
- Winged forms of adult aphids can travel large distances on winds
- Virus transmission can often also occur through mechanical transmission involved with plant propagation or management
- Production nursery greenhouses and significant proportions of Australia have favourable climatic conditions for both virus development and aphid spread and establishment

7.2.2 Surveys for early detection of an incursion in a production nursery

Points to consider in effectively monitoring aphid populations in commercial production nurseries are:

- Initial surveys using yellow sticky card traps or water pan traps to determine the species of aphid present. Yellow sticky card traps will determine the presence of aphids and other insect species such as thrips and aphids. It is recommended that traps be placed at a density of 1 trap per 300 to 400 plants positioning the trap bottom at canopy height. The traps should remain in place for one week
- The position of aphids on leaves depends on the aphid species and the crop. For example, Green peach aphids may uniformly scatter on the leaf, yet in lettuce populations start on the lower leaves and move up the plant, and in other crops may be found on the underside of the leaf. Because of their size, detection may be dependent on careful visual inspection, preferably supplemented by use of a hand lens magnifier
- Water pan traps are frequently used for monitoring aphid populations
- If aphids are detected, leaves infested with aphids (as many life stages as possible) should be collected for identification of the species

Points to consider in monitoring virus infected material in commercial production nurseries are:

- The host range of the potential virus incursion must be determined and hosts grouped into risk categories for transmission and expression of the disease (high, medium and low)
- Conditions under which transmission, amplification and expression of the disease must be determined to assess the likelihood of detection and reporting through general surveillance and to assist develop protocols for targeted surveillance
- Potential pathways for distribution of virus-infected material must be determined
- Depending on the virus, distribution of the virus in the plant may be irregular and plant material with most likely infection should be determined
- Depending on the virus incursion, host species in Australia are likely to be numerous and widely dispersed and may be present within relevant industries, nurseries as well as home gardens, landscape plantings and weeds
- Virologist expertise will be needed to determine diagnostic protocols and sampling requirements including the age of plant material to be sampled, time of year and the potential to bulk samples from plant species or cultivars

Important points to consider when developing early detection surveys are:

- Awareness information should be targeted at people who are in regular close contact with potential hosts in high risk areas or movement vectors (e.g. production nursery operators)
- Systematic and careful inspection of nursery crops and propagative plant material is essential to prevent introduction of an aphid transmitted virus and limit its spread within and from contaminated nurseries. Where possible, early detection of disease symptoms while at low levels, will provide the best chance of eradication
- An inspector must be trained to recognise a particular aphid vector and the virus symptoms and other similar disorders for comparison (see Section 5.2.3). A nursery layout map that includes approximate locations of target species will be required to develop a strategy for surveys. A survey map should include species and cultivar names, locations, approximate quantity and sources of targeted plants within the area. During the survey walkthrough, record the date, observations, and sampling information directly onto the survey map. The recorded information should be reviewed and used to develop an efficient survey strategy each time the nursery is inspected.

7.2.3 Delimiting surveys in the event of an incursion

- In the event of an incursion, delimiting surveys are essential to inform the decision-making process
- The size of the survey area will depend on the size of the infected area and the severity of the infection, as well as distribution pathways for plant material and potentially weather patterns during the period prior to detection (Figure 12). Other considerations are for example, movement of people or plant material equipment as a result of trace-forward and trace-backs
- Aphids can fly and can readily spread long distances by winds or can be transported on infested plants. New introductions can pose serious threats and complicate identification of naturalised populations

- All potential host species (refer to Section 5.2) should be surveyed, with particular attention paid to the species in which the virus was initially detected
- In addition to inspection of possible host plants, material should be collected for diagnostic purposes (refer to Section 7.2.4)
- If the incursion is in a populated area, publication and distribution of information sheets and appeals for public assistance may be helpful

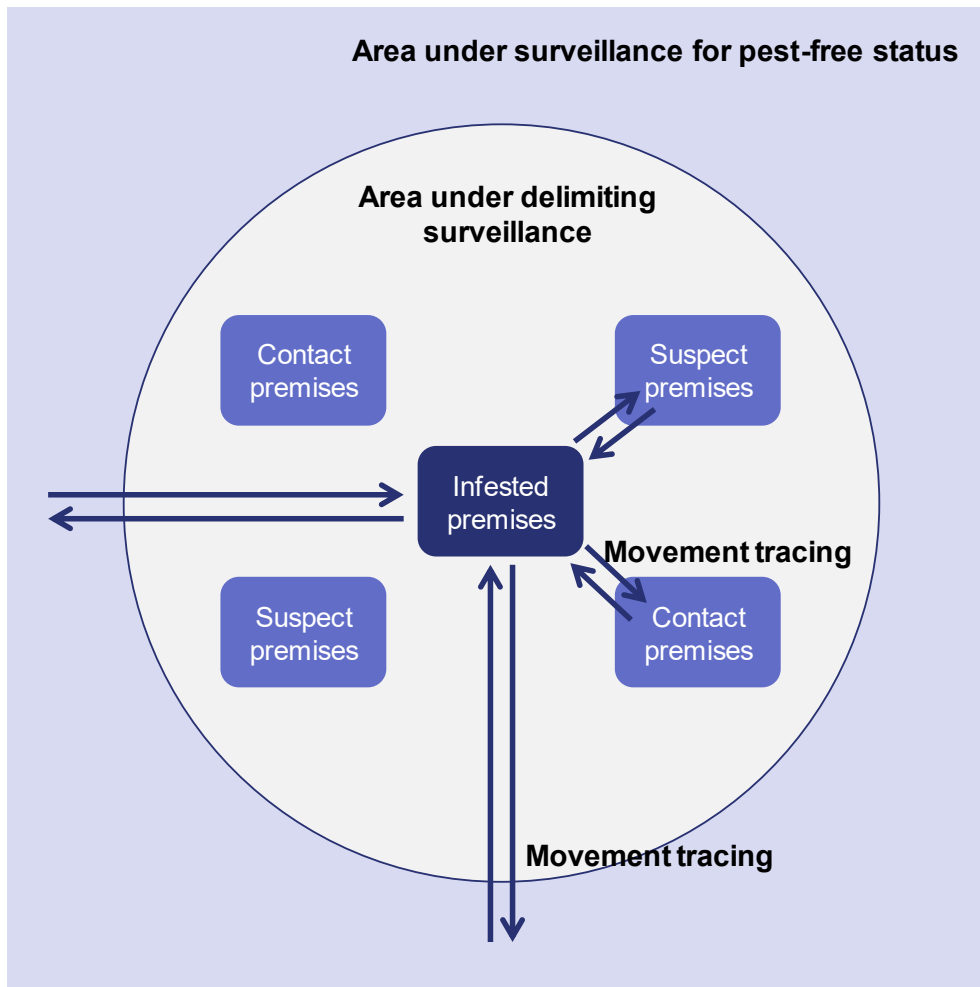


Figure 12. Diagram of a delimiting survey showing surveillance activities from the infected premises

7.2.4 Collection and treatment of aphid samples

Protocols for the collection, transport and diagnosis of suspect Emergency Plant Pests (EPPs) must follow PLANTPLAN (Plant Health Australia 2010). Any personnel collecting samples for assessment should notify the diagnostic laboratory prior to submitting samples to ensure expertise is available to undertake the diagnosis.

All sample containers should be clearly labelled with the name, address and contact phone number of both the sending and receiving officers. In addition containers should be clearly labelled in accordance with the requirements of PLANTPLAN (Plant Health Australia 2010). Containers should be carefully sealed to prevent loss, contamination or tampering of samples. The Chief Plant Health Manager will select the preferred laboratory. Additional labelling includes the identification of affected

plant species/parts, location of nursery and affected plant within the nursery (preferably with a GPS reading) as well as symptoms and an image if available.

Refer to PLANTPLAN for packing instructions under IATA 650.

7.2.4.1 COLLECTION OF SPECIMENS

Sampling procedures

Samples can be collected on leaf samples, yellow sticky traps, suction traps or water pan traps.

Number of specimens to be collected

Where possible, collect multiple specimens representative of all life stages of the population available. Hollingsworth and Gatsonis (1990) reported that a minimum of 10 and maximum of 50 leaf samples should be taken to monitor levels of *M. persicae* in field-grown potato.

Record the identity of the host plant where the aphids were collected. Record the location, preferably as GPS co-ordinates, or alternatively, a map reference or distance and direction from a suitable landmark. If the land is privately owned, record the owner's details including contact telephone numbers.

How to collect and send plant samples with eggs, larvae or pupae

Leaf samples containing nymphs and if possible adults are to be placed in a specimen container and placed in a portable fridge or insulated container with cool packs to prevent the insect and leaf samples from drying out.

All sample containers should be clearly labelled with the name, address and contact phone number of both the sending and receiving officers. In addition containers should be clearly labelled in accordance with the requirements of PLANTPLAN (Plant Health Australia 2010; Appendix 3). Containers should then be carefully sealed to prevent loss, contamination or tampering of samples. The Chief Plant Health Manager will select the preferred laboratory. Additional labelling includes the identification of plant species/parts affected, location of affected plant (where available include GPS reading) as well as symptoms and an image if available.

Refer to PLANTPLAN for packing instructions under IATA 650.

Precaution

Overheating or desiccation of samples prior to despatch should be prevented. Samples may be stored in a fridge (4-10°C) for a few days if necessary.

Receipt

On receipt of the samples the diagnostic laboratory should follow strict quarantine and processing guidelines. In keeping with ISO 17025 refer to PLANTPLAN (Plant Health Australia 2010).

7.2.5 Collection and treatment of virus samples

In general, plants showing virus like symptoms or suspected symptoms should be sampled. For PPV, when sampling leaf material, leaves from the middle of the branch should be sampled from various points around the tree. Optimal tissue for peach samples is petals if possible (National Diagnostic Protocol for Plum pox virus [PPV]). Any personnel collecting samples for assessment should notify the diagnostic laboratory prior to submitting samples to ensure expertise is available to undertake the diagnosis. General protocols for collecting and dispatching samples are available within Appendix 3 PLANTPLAN (Plant Health Australia 2010).

Number of specimens to be collected

In general, 5-10 samples of symptomatic plants should be collected for initial identification. If a survey to determine the incidence of disease within a crop or geographic area is required, then a more formalised, statistical-based sampling strategy should be employed.

For PPV, sampling protocols are listed in the National Diagnostic Protocol for Plum pox virus (PPV) and state that leaves of four trees will constitute a field sample, and 12-16 leaves will be collected per tree. Within the diagnostic test, a bulk sample of no more than 8 leaves will be tested.

It is important to record the precise location of all samples collected, preferably using GPS, or if this is not available, map references including longitude and latitude and road names should be recorded. Property and owners names should also be included where possible.

How to collect plant samples

Samples should be treated in a manner that allows them to arrive at the laboratory in a fresh, well-preserved state. An esky with ice packs or portable fridge should be carried when sampling crops.

The samples should be kept cool, out of direct sunlight and clearly labelled. Aim to keep the tissue at less than 10°C. For appropriate labelling and packaging procedures for suspect emergency plant pests consult PLANTPLAN (Plant Health Australia 2010).

Sampling and collection of plant material will depend on the host and virus. Technical advice on sampling will be required for each virus if an incursion is suspected.

In general, infected plant material should be collected using scissors, with sterilisation to occur between each collection.

For PPV, leaves should be collected into zip-lock plastic bags, with air removed and without paper towelling, or any material that will keep the plant leaves wet (National Diagnostic Protocol for Plum pox virus [PPV])

How to preserve plant samples

Collected material can be stored at 2-5°C. Do not expose plant samples to direct sunlight. It is important to keep the sampled plant tissue below 10°C where possible.

How to transport plant samples

Plant material should be mailed as a flat package. The samples should be either sent by a courier or by Express Post if overnight delivery to the diagnostic laboratory is guaranteed. Each laboratory has different labelling protocols and the receiving laboratory must be contacted before sending samples to

ensure that these protocols are followed and that someone will be present to receive the samples. Email is not sufficient as a sole method of contact.

7.2.6 Epidemiological study

The extent of infestation in a production nursery, on a property or within a region will depend on the initial population size of the virus introduction and also on populations of the aphid vector and whether conditions have been favourable for the pest to spread from the initial location. Sampling should be based upon the origins of the initial suspect sample(s). Factors to consider will be:

- The proximity of other susceptible plants to the initial infestation source, including both current and previous crops. This will include crops in the production nursery or on the property with the initial detection and those on neighbouring properties
- Machinery or vehicles that have been into the infested area or in close proximity to the infestation source
- The extent of human movements into and around the infested area. A possible link to the recent importation of plant material from other regions should also be considered
- The source of any production nursery stock propagation material
- If any other crops have been propagated from the same source and/or distributed from the affected production nurseries
- Depending on the temperature and environmental conditions aphids can have up to 20 generations per year
- Many vector- transmitted viruses can easily be spread by mechanical transmission in sap on contaminated machinery and equipment and also potentially by contact between plants

7.2.7 Models of spread potential

No models of spread potential have been developed for aphid transmitted viruses.

7.2.8 Pest Free Area guidelines

Determination of Pest Free Areas (PFAs) should be completed in accordance with the International Standards for Phytosanitary Measures (ISPMs) 8 and 10 (IPPC 1998a, 1999).

In the event of an incursion, specific guidelines for surveys and monitoring will be provided by the Consultative Committee on Emergency Plant Pests (CCEPP). General points to consider are:

- Design of a statistical delimiting survey for symptoms on host plants (see Section 7.2 for points to consider in the design)
- Plant sampling should be completed as described in the BioSecure *HACCP* manual (Nursery and Garden Industry Australia 2008), including monitoring processes (summarised in Table 7 and Table 8), indicator plants and weed monitoring
- Surveys should also consider alternative hosts (see Section 5.2.1) and not be limited to the primary infected host
- Information (including absence of the pest) should be recorded

Table 7. Summary of monitoring processes for protected production areas as described in BioSecure HACCP Guidelines. Further specific guidelines may be provided by a CCEPP

Wear protective clothing when handling suspect samples
Walk at random through the area in a zigzag pattern
Take at least 10 minutes to inspect 10-20 plants or plug trays per 100 m ² of production area
Inspect the tops and bottoms of leaves, looking for any direct evidence of insects
Inspect the entire plant if it has less than 6 leaves, or from larger plants select six leaves from all parts of the plant (upper, lower, middle) and examine them individually
Inspect the length of all stems and branches for insects and symptoms
During individual plant inspection, examine the underside of the foliage for the presence of whiteflies
If any plants show suspect symptoms or evidence of eggs or larvae (refer to Symptoms Section 5.2.3) take a sample (refer to Section 9.2.4) to be formally diagnosed
Check for a problem that has occurred regularly in the past, until you are certain it is not present
Record on the 'Crop Monitoring Record' sheet the presence or absence of the pest
Routinely inspect growing areas and remove alternate hosts and reservoirs of the pest, including weeds, crop residues and old plants that will not be marketed

Additional information is provided by the IPPC (1995) in Requirements for the Establishment of Pest Free Areas. This standard describes the requirements for the establishment and use of pest free areas as a risk management option for phytosanitary certification of plants and plant products. Establishment and maintenance of a PFA can vary according to the biology of the pest, pest survival potential, means of dispersal, availability of host plants, restrictions on movement of produce, as well as PFA characteristics (size, degree of isolation and ecological conditions).

Table 8. Summary of monitoring processes for field production areas as described in BioSecure HACCP Guidelines. Further specific guidelines may be provided by a CCEPP.

Wear protective clothing when handling suspect samples
Pay particular attention to areas on the windward side, the sides bordering ditches, canals or other uncultivated areas and growing block centres
Place a flag or other marker at the entrance to the block or sampling area at the beginning of each inspection
Vary the entrance point in the sampling area (1 m to 3 m) for each subsequent sampling so that the same plants are not inspected each time
Walk at random through the area in a zigzag pattern
The scout should follow the same general pattern at each sampling
Make an effort to select those plants that appear less healthy for visual inspection
Take at least 10 minutes to inspect 10-20 plants or plug trays per 100 m ² of production area
Inspect the tops and bottoms or leaves, looking for any direct evidence of insects
Inspect the entire plant if it has less than 6 leaves, or from larger plants select six leaves from all parts of the plant (upper, lower, middle) and examine them individually
Inspect the length of all stems and branches for insects and symptoms
During individual plant inspection, strike the foliage over a white sheet of paper, or a plastic or paper plate to dislodge small insects for easier viewing
If any plants show suspect symptoms or evidence of eggs or larvae (refer to Symptoms Section 5.2.3) take a sample (refer to Section 7.2.4) to be formally diagnosed
Check for a problem that has occurred regularly in the past, until you are certain it is not present
Record on the 'Crop Monitoring Record' sheet the presence or absence of the pest
Routinely inspect growing areas and remove alternate hosts and reservoirs of the pest, including weeds, crop residues and old plants that will not be marketed

7.3 Availability of control methods

7.3.1 General procedures for control

Control of aphid transmitted viruses is likely to be largely reliant on control measures for the aphid vectors. However, measures can also be taken to minimise the spread of virus-infected nursery stock. Specific control measures will be determined by a CCEPP, however, general procedures include:

- Keep traffic out of affected areas and minimise movement in adjacent areas
- Adopt best-practice property hygiene procedures to retard the spread of the pest between fields and adjacent properties
- After surveys are completed, and permission has been obtained from the Chief Plant Health Manager or the CCEPP, destruction of the infested plant material is an effective control
- On-going surveillance of infected areas to ensure the pest is eradicated
- Do not use any material from infected plants for propagation

For general management (i.e. not eradication), controlling aphid populations before they reach large numbers in crops is very important and requires close monitoring of aphid populations early in the

season. If the adults occur in large numbers it becomes difficult to control the nymphal stages. Adults move between successive crops, so management approaches must be employed in all crops within the area.

To reduce early season populations, best management practices require consideration of several management approaches including the use of pest-free seedlings, weed management, chemical control and cleaning up of crop residue. Winged Green peach aphids generally begin migrating from weeds to initiate colonies on crop seedlings in autumn in Australia, and populations peak in late winter and early spring.

It should be recognised that it will be extremely difficult for effective eradication of aphid populations and this will in turn limit the ability to eliminate all transmission of an aphid transmitted virus.

7.3.2 Pest free (clean) seedlings

Seedlings are potentially a major means of spreading aphids and aphid transmitted viruses into new plantings. Young plants are more susceptible to damage from aphids, so early infestations need to be avoided. Clean seedlings/nursery stock can be the first line of protection against the development of damaging populations.

Growers should check their suppliers to determine how the seedlings or nursery stock are grown and what measures are being used to protect against aphid infestation and virus infection. Inspect transplants carefully for signs of aphids or virus expression.

7.3.3 Weed management

Minimising aphid host plants is important in reducing the base aphid population at the start of the season, which will delay the time it takes for aphid numbers to reach significant levels, reducing the number of sprays needed for control. However, Green peach aphid (a major vector for TEV and PPV) feeds on hundreds of host plants in over 40 plant families, so controlling all host plants may not be an option.

Potviruses are typically non-persistent in the aphid vectors, so host plants of the vector that do not host the virus are not as great a concern as they are with persistent viruses. In the case of TEV for example, controlling host plants of the virus (such as tomato, capsicum, tobacco and solanaceous weeds) will limit the spread and survival of the virus.

7.3.4 Chemical control

Chemical control of viruses is not an option, but chemicals may be effective in trying to eliminate aphid vectors in an eradication campaign, or in the management of the aphid vectors should eradication be deemed unfeasible. In the event of an incursion of an aphid thrips transmitted virus, several chemicals, as listed on infopest, are currently registered for the control of aphids in Australia (Table 9).

Table 9. Registered chemicals and chemical use permits for the control of aphids in crops (from Infopest 2011)

Chemical group	Active ingredient	APVMA Permit	State	Use
	aldicarb 150 g kg ⁻¹	11105	NT	Aphid control in garlic seed production
	diafenthion 500 g kg ⁻¹	11971	Qld, NT, Vic, WA, SA, Tas, NSW	Aphid control in nursery stock (non-food) and fruit trees (non-food)
	diazinon 800 g L ⁻¹	12187	Tas	Black cherry aphid control on sweet cherries
	imidacloprid 200 g L ⁻¹	10497	Qld, NT, WA, SA, Tas, NSW	Aphid control in <i>Brassica</i> leafy vegetables
		9894	Qld, NT, WA, SA, Tas, NSW	Aphid control in rhubarb crops
		Registered: Agspray aphid guard	Qld, NT, Vic, WA, SA, Tas, NSW	Aphid control in certain vegetables and turf
	lambda-cyhalothrin 250 g L ⁻¹	11801	NT	Aphid control in garlic
	pirimicarb 500 g kg ⁻¹	11941	Qld, NT, WA, SA, Tas, NSW	Aphid control in eggplants and almonds
		11800	NT	Aphid control in garlic
		11763	Qld, NT, WA, SA, Tas, NSW	Aphid control in spring onions
		11439	Qld, NT, WA, SA, Tas, NSW	Aphid control in sweet potato, <i>Brassica</i> leafy vegetables, chicory and coriander
		10433	Qld, NT, WA, SA, Tas, NSW	Aphid control in sweet corn
		8613	Qld, NT, WA, SA, Tas, NSW	Aphid control in leafy and woody herbs as specified when grown as annuals
		Registered: Farmoz aphidex	Qld, NT, Vic, WA, SA, Tas, NSW	Control of certain aphids on crops and pastures

Chemical group	Active ingredient	APVMA Permit	State	Use
	pirimicarb 500 g kg ⁻¹ + pyrethrins 75 g kg ⁻¹	11292	Tas	Aphid control in pyrethrum (restricted to approved persons)
	pirimicarb 500 g L ⁻¹	11194	Qld, NT	Control of Cowpea aphid and Soybean aphid in adzuki bean, mung bean and soybean
	Pymetrozine 500 g kg ⁻¹	11988	Qld, NT, WA, SA, Tas, NSW	Control of Lettuce aphid in specified vegetable crops
		11973	Qld, NT, Vic, WA, SA, Tas, NSW	Aphid control in nursery plants (non-food), seedlings and plugs
		10005	Qld, NT, WA, SA, Tas, NSW	Control of Green peach aphid in almonds
		9317	Qld, NT, WA, SA, Tas, NSW	Aphid control in snow peas and sugar snap peas
	Spirotetramat 240 g L ⁻¹	11839	Qld, NT	Control of Green peach aphid in seed crops of maize and sorghum
		11775	Qld, NT, WA	Aphid control in seed crops of maize, sorghum and sunflower

7.3.5 Clean-up crop residues

For control of aphids, movement of adults from older crops and crop residues is the main source of local infestation for younger crops. Post-harvest destruction of heavily infested crops often causes mass migration of aphids into adjacent crops. Therefore it is important to control aphids before they move into young crops.

Clean-up strategies for aphid infested crops/crop residues:

- For moderate aphid infestations, use an insecticide effective against adults
- Use high spray volumes for better coverage as defined on the label
- Re-entry/withholding periods still apply. Entry into crops should be avoided and produce should not be taken from the fields for consumption. Crop residues should not be fed to livestock
- Residues should be deep-buried or contained (sealed plastic bags) and disposed of via incineration

Clean up of virus-infected plant material could include:

- Spraying of host material (including newly emerging plants) with herbicide to kill the plants *in situ*, followed by disposal of plant material by deep burial
- Break down of larger volumes of plant material under black plastic prior to deep burial, (methodology to be confirmed by CCEPP)
- Burning of plant material providing
 - Material is sufficiently dry to burn
 - Aphid populations have been controlled/eliminated to ensure they are not distributed in rising thermals or surrounding areas by burning
- Decontamination of all items in contact with decomposing plant material with a 1% a.i. sodium hypochlorite solution or by deep burial
- Keeping infested area free of hosts for a minimum of 12 months (or a period to be determined by a CCEPP)

7.3.6 Cultural Control

The use of sawdust, woodchips or mulches (organic or aluminium) can reduce aphid visits to plants and thus reduce spread of some viruses (e.g. TEV, AVRDC 2004). Aphid trap crops (such as beans) surrounding field plantings of capsicums can be effective in reducing TEV spread provided that aphids are chemically controlled within the trap crop (Sherf and MacNab 1986).

The use of aphid-proof glasshouses or greenhouses for raising seedlings will strongly reduce the chance of virus infection.

7.3.7 Host-Plant Resistance

For TEV, resistant capsicum/pepper varieties are available but there are no resistant tomato varieties at present.

For PPV, the natural hosts are restricted to *Prunus* and it should be noted that not all PPV isolates infect all *Prunus* species. Despite significant efforts to identify resistance to PPV, few reliable sources of high level resistance have been found. Resistance to PPV has been identified in the model plant species *Arabidopsis thaliana* (Decroocq *et al.* 2006). High levels of resistance have also been produced to the D and M strains using genetic engineering in the transgenic C5 plum (Levy *et al.* 2000). In addition, multigenic sources of resistance have been shown to provide moderate levels of resistance or tolerance to some strains of PPV (Kegler *et al.* 1998) however incorporation of multiple genes, especially in combination with quality, yield and agronomic traits makes breeding difficult.

7.3.8 Insect Pest Management

The use of mineral oil sprays can delay virus spread by interfering with aphid transmission of viruses (AVRDC 2004).

There are many natural enemies of *M. persicae*, some of which are specific and others that are general to all aphids. These include syphid maggots, *Allograpta* sp., lady beetles and parasitic wasps. Another effective parasite is *Diaretus chenopodiaphidis* Ashmead. Parasites of *A. spiraecola* include wasps of the genera *Aphidius* and *Aphelinus*, and predators including hoverfly larvae, lacewing larvae and ladybird beetles (Broughton 2007).

7.3.9 Managing viruses

The two key points in managing the spread of TEV and PPV virus are to:

- Prevent the movement of infected host plants, seedlings and aphid-infected plants
- Control aphids on-farm, in surrounding vegetation and in seedling nurseries using good farm management and farm hygiene practices
- Minimise handling during the growing season to reduce the mechanical spread of TEV (AVRDC 2005)
- Use virus free nursery stock

7.4 Phytosanitary Measures

There are no known phytosanitary measures for *S. persicae* or *A. spiraecola* because they already have a worldwide distribution.

Because of the difficulty of detecting low levels of infestation in consignments, it is best to ensure that the place of production is free from the pest (OEPP/EPPO, 1990). Particular attention is needed for consignments from countries where aphid-transmitted viruses are present.

Of the list provided in Table 3, EPPO lists Plum pox virus as an A2 quarantine pest (i.e. present in Europe but with limited distribution).

8 Course of action

Additional information is provided by the IPPC (1998b) in Guidelines for Pest Eradication Programmes. This standard describes the components of a pest eradication programme which can lead to the establishment or re-establishment of pest absence in an area. A pest eradication programme may be developed as an emergency measure to prevent establishment and/or spread of a pest following its recent entry (re-establish a pest free area) or a measure to eliminate an established pest (establish a pest free area). The eradication process involves three main activities: surveillance, containment, and treatment and/or control measures.

8.1 Destruction strategy

8.1.1 Destruction protocols

- General protocols:
 - No plant material should be removed from the infested area unless part of the disposal procedure
 - Disposable equipment, infested plant material or growing media/soil should be disposed of by autoclaving, high temperature incineration or deep burial
 - Any equipment removed from the site for disposal should be double-bagged
 - Machinery used in destruction processes need to be thoroughly washed, preferably using a detergent or farm degreaser

8.1.2 Decontamination protocols

Machinery, equipment and vehicles in contact with infested plant material or growing media/soil, or present within the Quarantine Area, should be washed to remove plant material and growing media/soil using high pressure water or scrubbing with products such as a degreaser or a bleach solution (1% available chlorine) in a designated wash down area. When using high pressure water, care should be taken not to spread plant material. High pressure water should be used in wash down areas which meet the following guidelines:

- Located away from crops or sensitive vegetation
- Readily accessible with clear signage
- Access to fresh water and power
- Mud free, including entry and exit points (e.g. gravel, concrete or rubber matting)
- Gently sloped to drain effluent away
- Effluent must not enter water courses or water bodies
- Allow adequate space to move larger vehicles
- Away from hazards such as power lines
- Waste water, growing media/soil or plant residues should be contained (see Appendix 18 of PLANTPLAN [Plant Health Australia 2010])

- Disposable overalls and rubber boots should be worn when handling infested plant material or growing media/soil in the field. Boots, clothes and shoes in contact with infested plant material or growing media/soil should be disinfected at the site or double-bagged to remove for cleaning
- Skin and hair in contact with infested plant material or growing media/soil should be washed

Procedures for the sterilisation of plant containers and growing media are provided within the BioSecure HACCP Guidelines, however, in the event of an aphid transmitted virus incursion, additional or modified procedures may be required for the destruction of the pest. Any sterilisation procedure must be approved for use in the endorsed Response Plan.

8.1.3 Priorities

- Confirm the presence of the pest
- Limit movement of people and prevent movement of vehicles and equipment through affected areas
- Stop the movement of any plant material that may be infested with the pest
- Determine the strategy for the eradication/decontamination of the pest and infested host material
- Determine the extent of infestation through survey and plant material trace back and trace forward which would be assessed on a case by case basis and included within the response plan

8.1.4 Plants, by-products and waste processing

- Any growing media/soil or infected plant material removed from the infected site should be destroyed by (enclosed) high temperature incineration, autoclaving or deep burial
- As aphid transmitted viruses are easily spread, plant debris from the destruction zone must be carefully handled and transported
- Infested areas or production nursery property should remain free of susceptible host plants until the area has been shown to be free from the pathogen

8.1.5 Disposal issues

- Particular care must be taken to minimise the transfer of infected plant material from the area
- Host material including leaf litter should be collected and incinerated or double bagged and deep buried in an approved site

8.2 Containment strategies

For some exotic pest incursions where eradication is considered impractical, containment of the pest may be attempted to prevent or slow its spread and to limit its impact on other parts of the state or country. Containment is currently being considered for inclusion within the Emergency Plant Pest

Response Deed (EPPRD). The decision on whether to eradicate or contain the pest will be made by the National Management Group, based on scientific and economic advice. Emergency interim containment measures are possible under EPPRD arrangements to gather information to determine if eradication is technically feasible.

8.3 Quarantine and movement controls

Consult PLANTPLAN (Plant Health Australia 2010) for administrative details and procedures.

8.3.1 Quarantine priorities

- Plant material and growing media/soil at the site of infestation to be subject to movement restrictions
- Machinery, equipment, vehicles and disposable equipment in contact with infested plant material or growing media/soil, or present in close proximity to the site of infestation to be subject to movement restrictions

8.3.2 Movement controls

Movement controls need to be put in place to minimise the potential for transport of the pest, and this will apply to all plant material, growing media and other items within the quarantined area.

Movement of people, vehicles, equipment and plant material, from and to affected properties or areas, must be controlled to ensure that the pest is not moved off-property. Movement controls can be achieved through the following, however specific measures must be endorsed in the Response Plan:

- Signage to indicate quarantine area and restricted movement into and within these zones
- Fenced, barricaded or locked entry to quarantine areas
- Movement of equipment, machinery, plant material or growing media/soil by permit only. Therefore, all non-essential operations in the area or on the property should cease
- Where no dwellings are located within these areas, strong movement controls should be enforced
- Where dwellings and places of business are included within the Restricted and Control Areas movement restrictions are more difficult to enforce, however limitation of contact with infested plants should be enforced
- If a production nursery is situated within the Restricted Area, all trading in host and non-host material must cease and no material should be removed from the site without permission, due to the high likelihood of pest spread. Movement restrictions would be imposed on both host and non-host material
- Residents should be advised on measures to minimise the inadvertent transport of aphids from the infested area to unaffected areas
- Clothing and footwear worn at the infested site should either be double-bagged prior to removal for decontamination or should not leave the site until thoroughly disinfected, washed and cleaned

- Plant material or plant products must not be removed from the site unless part of an approved disposal procedure.
- All machinery and equipment should be thoroughly cleaned down with a high pressure cleaner (see Section 8.1.2) or scrubbing with products such as a farm degreaser or a 1% bleach (available chlorine) solution, prior to leaving the affected area. Machinery should be inspected for the presence of insects and if found, treatment with insecticide may be required. The clean down procedure should be carried out on a hard surface, preferably a designated wash-down area, to avoid mud being re-collected from the affected site onto the machine. When using high pressure water, care should be taken to contain all plant material and mud dislodged during the cleaning process

8.4 Zoning

The size of each quarantine area will be determined by a number of factors, including the location of the incursion, biology of the pest, climatic conditions and the proximity of the infested property to other infested properties. This will be determined by the National Management Group during the production of the Response Plan. Further information on quarantine zones in an Emergency Plant Pest (EPP) incursion can be found in Appendix 10 of PLANTPLAN (Plant Health Australia 2010). These zones are outlined below and in Figure 13.

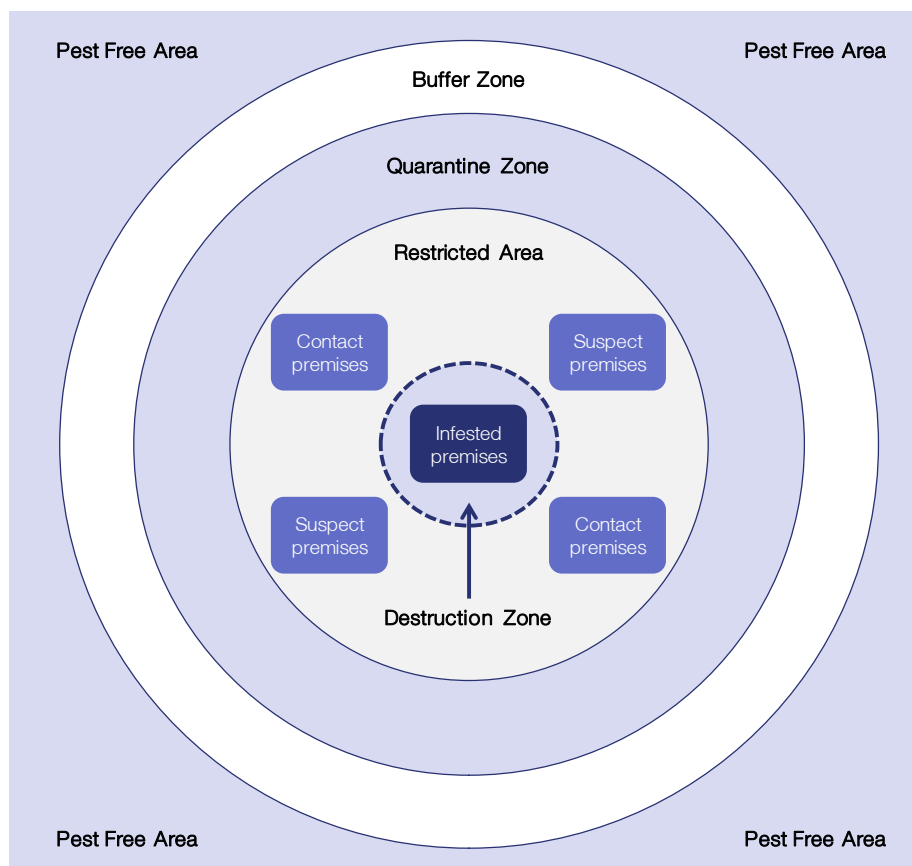


Figure 13. Schematic diagram of quarantine zones used during an EPP incursion (not drawn to scale)

8.4.1 Destruction Zone

The size of the destruction zone (i.e. zone in which the pest and all host material is destroyed) will depend on the ability of the pest to spread, distribution of the pest (as determined by delimiting surveys), time of season (and part of the pest life cycle being targeted) and factors which may contribute to the pest spreading.

All host plants should be destroyed after the level of infestation has been established. The delimiting survey will determine whether or not neighbouring plants are infested and need to be destroyed. Non-host plant material within this zone may be destroyed, based on recommendations in the Response Plan. The Destruction Zone may be defined as contiguous areas associated with the same management practices as, or in contact with, the infested area (i.e. the entire production nursery, property or area if spread could have occurred prior to the infection being identified). It should also consider the predicted range associated with movement of the vector.

Particular care needs to be taken to ensure that plant material (including non-hosts) is not moved into surrounding areas.

8.4.2 Quarantine Zone

The Quarantine Zone is defined as the area where voluntary or compulsory restraints are in place for the affected property or properties. These restraints may include restrictions or movement control for removal of plants, people, growing media/soil or contaminated equipment from an infected property.

8.4.3 Buffer Zone

A Buffer Zone may or may not be required depending on the incident. It is defined as the area in which the pest does not occur but where movement controls or restrictions for removal of plants, people, soil or equipment from this area are still deemed necessary. The Buffer Zone may enclose an infested area (and is therefore part of the Control Area) or may be adjacent to an infested area.

8.4.4 Restricted Area

The Restricted Area is defined as the zone immediately around the infected premises and suspected infected premises. The Restricted Area is established following initial surveys that confirm the presence of the pest. The Restricted Area will be subject to intense surveillance and movement control with movement out of the Restricted Area to be prohibited and movement into the Restricted Area to occur by permit only. Multiple Restricted Areas may be required within a Control Area.

8.4.5 Control Area

The Control Area is defined as all areas affected within the incursion. The Control Area comprises the Restricted Area, all infected premises and all suspected infected premises and will be defined as the minimum area necessary to prevent spread of the pest from the Quarantine Zone. The Control Area will also be used to regulate movement of all susceptible plant species to allow trace back, trace forward and epidemiological studies to be completed.

8.5 Decontamination and farm clean up

Decontaminant practices are aimed at eliminating the pathogen thus preventing its spread to other areas.

8.5.1 Decontamination procedures

General guidelines for decontamination and clean up:

- Refer to PLANTPLAN (Plant Health Australia 2010) for further information
- Keep traffic out of affected area and minimise it in adjacent areas
- Adopt best-practice property hygiene procedures to retard the spread of the pest between growing areas/fields and adjacent properties
- Machinery, equipment, vehicles in contact with infested or infected plant material or growing media/soil present within the Quarantine Zone, should be washed to remove growing media/soil and plant material using high pressure water or scrubbing with products such as a degreaser or a bleach solution in a designated wash down area as described in Section 8.1.2
- Only recommended materials are to be used when conducting decontamination procedures, and should be applied according to the product label
- Infested plant material should be disposed of by autoclaving, high temperature (enclosed) incineration or deep burial

8.5.2 General safety precautions

For any chemicals used in the decontamination, follow all safety procedures listed within each MSDS.

8.6 Surveillance and tracing

8.6.1 Surveillance

Detection and delimiting surveys are required to delimit the extent of the outbreak, ensuring areas free of the pest retain market access and appropriate quarantine zones are established.

Initial surveillance priorities include the following:

- Surveying all host growing properties and businesses in the pest quarantine area
- Surveying all properties and businesses identified in trace-forward or trace-back analysis as being at risk
- Surveying all host growing properties and businesses that are reliant on trade with interstate or international markets which may be sensitive to pathogen presence
- Surveying production nurseries selling at risk host plants
- Surveying other host growing properties and backyards

8.6.2 Survey regions

Establish survey regions around the surveillance priorities identified above. These regions will be generated based on the zoning requirements (see Section 8.4), and prioritised based on their potential likelihood to currently have or receive an incursion of this pest. Surveillance activities within these regions will either allow for the area to be declared pest free and maintain market access requirements or establish the impact and spread of the incursion to allow for effective control and containment measures to be carried out. Detailed information regarding surveys for aphid and virus infected plant material have been outlined elsewhere in this plan (refer to Section 7.2).

Steps outlined in Table 10 form a basis for a survey plan. Although categorised in stages, some stages may be undertaken concurrently based on available skill sets, resources and priorities.

Table 10. *Phases to be covered in a survey plan*

Phase 1	Identify properties that fall within the buffer zone around the infested premise Complete preliminary surveillance to determine ownership, property details, production dynamics and tracings information (this may be an ongoing action)
Phase 2	Preliminary survey of host crops in properties in buffer zone establishing points of pest detection
Phase 3	Surveillance of an intensive nature, to support control and containment activities around points of pest detection
Phase 4	Surveillance of contact premises. A contact premise is a property containing susceptible host plants, which are known to have been in direct or indirect contact with an infested premises or infected plants. Contact premises may be determined through tracking movement of materials from the property that may provide a viable pathway for spread of the disease. Pathways to be considered are: <ul style="list-style-type: none"> • Movement of plant material and growing media/soil from controlled and restricted areas • Items of equipment and machinery which have been shared between properties including bins, containers, irrigation lines, vehicles and equipment • The producer and retailer of infected material if this is suspected to be the source of the outbreak • Labour and other personnel that have moved from infected, contact and suspect premises to unaffected properties (other growers, tradesmen, visitors, salesmen, crop scouts, harvesters and possibly beekeepers) • Storm and rain events and the direction of prevailing winds that result in air-borne dispersal of the pathogen during these weather events
Phase 5	Surveillance of production and greenlife retailers, including garden centres, hardware outlets and supermarkets, as well as gardens and public land where plants known to be hosts of pathogen are being grown
Phase 6	Agreed area freedom maintenance, post control and containment

8.6.3 Post-eradication surveillance

The period of pest freedom sufficient to indicate that eradication of the pest has been achieved will be determined by a number of factors, including growth conditions, the previous level of infection, the control measures applied and the pest biology.

Specific methods to confirm eradication of aphid transmitted viruses may include:

- Monitoring of sentinel plants

- Sentinel plants are to be grown in containers or small plots at the affected site. Plants are to be grown *in situ* under quarantine conditions and monitored for symptoms of infection
- If symptoms or virus are detected, samples are to be collected and stored and plants destroyed
- Surveys comprising host plant sampling for the virus should be undertaken for a minimum of three years after eradication has been achieved (or as endorsed by a CCEPP)

9 Technical debrief and analysis for stand down

Refer to PLANTPLAN (Plant Health Australia 2010) for further details

The emergency response is considered to be ended when either:

- Eradication has been deemed successful by the lead agency, with agreement by the Consultative Committee on Emergency Plant Pests and the Domestic Quarantine and Market Access Working Group
- Eradication has been deemed impractical and procedures for long-term management of the disease risk have been implemented

A final report should be completed by the lead agency and the handling of the incident reviewed.

Eradication will be deemed impractical if, at any stage, the results of the delimiting surveys lead to a decision to move to containment/control.

10 References

Avinent L, Hermoso de Mendoza A, Llacer G (1994) Transmission of plum pox potyvirus in Spain. OEPP Bulletin 24, 669-674.

AVRDC (2004) Pepper diseases: Tobacco etch virus (TEV). AVRDC -The World Vegetable Centre - Fact sheet, Taiwan. www.avrdc.org

AVRDC (2005) Tomato diseases: Tobacco etch virus (TEV). AVRDC -The World Vegetable Centre - Fact sheet, Taiwan. www.avrdc.org

Brault V, Uzest M, Monsion B, Jacquot E, Blanc S (2010) Aphids as transport devices for plant viruses. *Virology* 333, 524-538.

Broughton S (2007) Aphids in citrus. Department of Agriculture and Food Western Australia Farmnote. www.agric.wa.gov.au

Cambra M, Capote N, Myrta A, Ll  cer G (2006) Plum pox virus and the estimated costs associated with sharka disease. Bulletin OEPP/EPPO Bulletin, 36(2):202-204.

Candresse T, Cambra M (2006) Causal agent of sharka disease: historical perspective and current status of Plum pox virus strains. OEPP/EPPO Bulletin 36, 239-246.

Candresse T, Dosba F, Quiot JB, Dunez J (1993) Sharka. The state of research. *Arboriculture Fruiti  re* 464, 30-35.

- Carrasco P, Daròs JA, Agudelo-Romero P, Elena SF (2007) A real-time RT-PCR assay for quantifying the fitness of tobacco etch virus in competition experiments. *Journal of Virological Methods* 139, 181-188.
- Cook AA, Ozaki HY, Zitter TA, Blazquez CH (1976) Florida VR-2, a bell pepper with resistances to three virus diseases. Florida Agricultural Experiment Station Circular S-242.
- Danci O, Ziegler A, Torrance L, Gasemi S, Danci M (2009) Potyviridae family – short review. *Journal of Horticulture, Forestry and Biotechnology* 13, 410-420.
- Debrot EA (1976) Studies on tobacco etch virus in tomato plantations in Venezuela. *Agronomia Tropical*, 26, 321-335.
- Decroocq V, Sicard O, Alamillo JM, Lansac M, Eyquard JP, Garcia JA, Candresse T, Le Gall O, Revers F (2006) Multiple resistance traits control Plum pox virus infection in *Arabidopsis thaliana*. *Molecular Plant Microbe Interactions*, 19, 541-549.
- Diekmann M, Putter CAJ (1996) FAO/IPGRI technical guidelines for the safe movement of germplasm No. 15: *Musa* spp.
- Dosba F, Lansac M, POcheur G, Teyssier B, Piquemal JP, Michel M (1986) Plum pox virus detection by ELISA technique in peach and apricot infected trees at different growing stage. *Acta Horticulturae* 193, 187-191.
- Dunez J (1987) Plum Pox Disease of Stone Fruit Trees. Rome, Italy: FAO.
- Gibbs AJ, Mackenzie AM, Wei KJ, Gibbs MJ (2008) The potyviruses of Australia. *Archives of Virology* 153, 1411-1420.
- Gildow F, Damsteegt V, Stone A, Schneider W, Luster D, Levy L (2004) Plum pox in North America: identification of aphid vectors and a potential role for fruit in virus spread. *Phytopathology*, 94, 868-874.
- Glasa M, Palkovics L, Komínek P, Labonne G, Pittnerová S, Kúdela O, Candresse T, Šubr Z (2004) Geographically and temporally distant natural recombinant isolates of Plum pox virus (PPV) are genetically very similar and form a unique PPV subgroup. *Journal of General Virology*, 85, 2671-2681.
- Gottwald TR, Avinent L, Llacer G, Mendoza AH de, Cambra M (1995) Analysis of the spatial spread of sharka (plum pox virus) in apricot and peach orchards in eastern Spain. *Plant Disease* 79, 266-278.
- Greenleaf WH (1953) Effects of tobacco-etch virus on peppers (*Capsicum* sp.). *Phytopathology* 43, 564-570.
- Hollingsworth CS, Gatsonis CA (1990) Sequential sampling plans for green peach aphid (Homoptera: Aphididae) on potato. *Journal of Economic Entomology* 83, 1365-1369.
- IPPC (1995) Requirements for the Establishment of Pest Free Areas. International Standards for Phytosanitary Measures (ISPM) No. 4.
- IPPC (1998a) Determination of pest free status in an area. International Standards for Phytosanitary Measures (ISPM) No. 8.
- IPPC (1998b) Guidelines for Pest Eradication Programmes. International Standards for Phytosanitary Measures (ISPM) No. 9.
- IPPC (1999) Requirements for the establishment of pest free places for production and pest free production sites (ISPM) No.10.
- James D, Glasa M (2006) Causal agent of sharka disease: new and emerging events associated with Plum pox virus characterization. OEPP/EPPO Bulletin, 36, 247-250.

- James D, Varga A (2005) Nucleotide sequence analysis of Plum pox virus isolate W3174: evidence of a new strain. *Virus Research* 110, 143-150.
- James D, Varga A, Thompson D, Hayes S (2003) Detection of a new and unusual isolate of Plum pox virus in plum (*Prunus domestica*). *Plant Disease* 87, 1119-1124.
- Johnson EM (1930) Virus diseases of tobacco in Kentucky. Kentucky Agricultural Experiment Station Research Bulletin 306.
- Kalashyan YA, Bilkey ND, Verderevskaya TD, Rubina EV (1994) Plum pox potyvirus on sour cherry in Moldova. OEPP/EPPO Bulletin 24, 645-649.
- Kegler H, Hartmann W (1998) Present status of controlling conventional strains of plum pox virus. In: Hadidi A, Khetarpal RK, Koganezawa H, eds. *Plant Virus Disease Control*. St Paul, Minnesota, USA: APS Press, 616-628.
- Kegler H, Fuchs E, Grüntzig M, Schwarz S (1998) Some results of 50 years of research on the resistance to plum pox virus. *Acta Virologica*. 42, 200-215.
- Kerlan C, Dunez J (1979) Biological and serological differentiation of strains of sharka virus. *Annales de Phytopathologie* 11, 241-250.
- Kölber M, Nemeth M, TSkés G, Krizbai L, SzSnyegi S, Imber I, Bereczki Zs, Papp E, Kiss E, Imre P, Pocsai E, Hangyál R, Vollent, Pete A, Takács M, Bencze E, Hajnóczy GY, Mer SF (1997) Five-year study to determine of eventual occurrence of plum pox virus in cherry cultivars in Hungary. *Acta Horticulturae* 472, 495-502.
- Komazaki S (1993) Biology and virus transmission of citrus aphids. Technical Bulletin, ASPAC Food and Fertilizer Technology Centre, Taipei.
- Kunze L, Krczal H (1971) Transmission of sharka virus by aphids. In: *Proceedings of the 8th European Symposium on Fruit Tree Virus Diseases*. Paris, France: INRA, 255-260.
- Labonne G, Lauriaut F, Yvon M, Quiot JB (1994) Aphid transmission of plum pox potyvirus: analysis of potential vectors in an apricot orchard. OEPP/EPPO Bulletin 24, 681-690.
- Leclant F (1973) Ecological aspects of the transmission of Sharka disease (Plum pox) in south-eastern France. Discovery of new species of aphid vectors. *Annales de Phytopathologie*, 5, 431-439.
- Levy L, Damsteegt V, Scorza R, Kolber M (2000) Plum pox – potyvirus disease of stone fruits. The American Phytopathological Society
<http://www.apsnet.org/publications/apsnetfeatures/Pages/PlumPoxPotyvirus.aspx>
- Llácer G, Cambra M (2006) Hosts and symptoms of Plum pox virus: fruiting *Prunus* species. OEPP/EPPO Bulletin 36, 219-221.
- Llácer G, Cambra M, Lavina M, Arambura J (1986) Investigation on plum pox (sharka) virus in Spain. *Acta Horticulturae* 193, 155-158.
- Mau RFL, Kessing JLM (1991) *Myzus persicae* (Sulzer). University of Hawaii Extension Bulletin. <http://www.extento.hawaii.edu/kbase/crop/type/myzus.htm>
- McLean DM (1962) Tobacco-etch virus on peppers and tomatoes in the Lower Rio Grande valley of Texas. *Plant Disease Reporter* 46, 565-568.
- Merriman P, McKirdy S (2005) Technical guidelines for development of pest specific response plans. Plant Health Australia, Deakin, ACT.
- Nault LR (1997) Arthropod transmission of plant viruses – a new synthesis. *Annals of the Entomological Society of America* 90, 395-403.

Ng JCK, Perry KL (2004) Transmission of plant viruses by aphids. *Molecular Plant Pathology* 5, 505-511.

Nursery and Garden Industry Australia (2008) Biosecure HACPP: guidelines for managing biosecurity in nursery production.

OEPP/EPPO (1983) Data sheets on quarantine organisms No. 96, Plum pox virus. OEPP/EPPO Bulletin 13.

OEPP/EPPO (1990) Specific quarantine requirements. EPPO Technical Documents, No. 1008. Paris, France: EPPO.

Plant Health Australia (2010). PLANTPLAN Australian Emergency Plant Pest Response Plan. Version 1. (www.planthealthaustralia.com.au/plantplan).

Purcifull DE, Hiebert E (1982) Tobacco etch virus. CMI/AAB Descriptions of Plant Viruses, No. 258 (no. 55 revised). Wellesbourne, UK: Association of Applied Biologists, 7 pp.

Salvador B, García JA, Simón-Mateo C (2006) Causal agent of sharka disease: Plum pox virus genome and function of gene products. OEPP/EPPO Bulletin 36, 229-238.

Shepherd RJ, Purcifull DE (1971) Tobacco etch virus. CMI/AAB Descriptions of Plant Viruses, No. 55. Wellesbourne, UK: Association of Applied Biologists, 4 pp.

Sherf AF, Macnab AA (1986) Vegetable diseases and their control. John Wiley, New York.

Stover RH (1951) Association in tobacco of the severe symptom response to etch virus and the White Burley character. *Phytopathology* 41, 1125-1126.

SubCommittee for Plant Health Diagnostic Standards (SPHDS) National Diagnostic Protocol for *Plum pox virus* (PPV), protocol number NDP 2, endorsed 18 February 2010.

Toba HH (1964) Life-history studies of *Myzus persicae* in Hawaii. *Journal of Economic Entomology* 57, 290-291.

Wang JJ, Tsai JH (2000) Effect of temperature on the biology of *Aphis spiraecola* (Homoptera: Aphididae). *Annals of the Entomological Society of America* 93, 874-883.

Wetzel T, Candresse T, Ravelonandro M, Delbos RP, Mazyad H, Aboul-Ata AE, Dunez J (1991a) Nucleotide sequence of the 3'-terminal region of the RNA of the E1 Amar strain of plum pox potyvirus. *Journal of General Virology* 72, 1741-1746

Wetzel T, Candresse T, Ravelonandro M, Dunez J (1991b) A polymerase chain reaction assay adapted to plum pox potyvirus detection. *Journal of Virological Methods* 33, 355-365.

Zitter TA (1971) Virus diseases of pepper in South Florida. *Proceedings of the Florida State Horticultural Society* 84, 177-183.

Zitter TA, Ozaki HY (1973) Reaction of susceptible and tolerant pepper varieties to the pepper virus complex in South Florida. *Proceedings of the Florida State Horticultural Society* 86, 146-152.

Zitter TA, Tsai JH (1981) Viruses infecting tomato in southern Florida. *Plant Disease* 65, 787-791.

Websites

Agdia website www.agdia.com/

Center for Invasive Species and Ecosystem Health - Bugwood network www.bugwood.org

CABI 2011 www.cabicompendium.org/cpc/home.asp

EPPO archives archives.eppo.org/EPPOReporting/1998/Rse-9805.pdf

EPPO website

www.eppo.org/QUARANTINE/virus/Cucurbit_yellow_stunting_disorder/DS_cucurbit_yellow_stunting.pdf

IPPC website www.ippc.int

TEV diagnostics <http://www.srbiolab.com/>

11 Appendices

11.1 Appendix 1: Hosts of Green peach aphid (*Myzus persicae*) as listed on the CAB Compendium (CAB International 2011)

Species	Species
<i>Abelmoschus esculentus</i> (okra)	<i>Hordeum vulgare</i> (barley)
<i>Aleurites</i>	<i>Humulus lupulus</i> (hop)
<i>Aloe</i> (grey alder)	<i>Impatiens</i> (balsam)
<i>Anchusa</i> (Bugloss)	<i>Indigofera</i> (indigo)
<i>Anethum graveolens</i> (dill)	<i>Ipomoea batatas</i> (sweet potato)
<i>Anthriscus</i> (chervil)	<i>Iris</i> (irises)
<i>Antirrhinum</i> (snapdragon)	<i>Lactuca sativa</i> (lettuce)
<i>Apium graveolens</i> (celery)	<i>Lavandula angustifolia</i> (lavender)
<i>Araceae</i>	<i>Lepidium sativum</i> (garden cress)
<i>Arachis hypogaea</i> (groundnut)	<i>Lepidium virginicum</i> (Virginian peppergrass)
<i>Armoracia rusticana</i> (horseradish)	<i>Lilium</i> (lily)
<i>Artemisia</i> (wormwoods)	<i>Lolium</i> (ryegrasses)
<i>Asparagus officinalis</i> (asparagus)	<i>Lolium multiflorum</i> (Italian ryegrass)
<i>Beta vulgaris</i> var. <i>saccharifera</i> (sugarbeet)	<i>Lupinus</i> (lupins)
<i>Brassica</i>	<i>Malus domestica</i> (apple)
<i>Brassica oleracea</i> (cabbages, cauliflowers)	<i>Malva</i> (mallow)
<i>Brassica rapa</i> cultivar group Caixin	<i>Matthiola</i>
<i>Brassica rapa</i> subsp. <i>chinensis</i> (Chinese cabbage)	<i>Medicago sativa</i> (lucerne)
<i>Cajanus cajan</i> (pigeon pea)	<i>Mentha</i> (mints)
<i>Capsella bursa-pastoris</i> (shepherd's purse)	<i>Narcissus</i> (daffodil)
<i>Capsicum</i> (peppers)	<i>Papaver somniferum</i> (Opium poppy)
<i>Capsicum annuum</i> (bell pepper)	<i>Pastinaca sativa</i> (parsnip)
<i>Carica papaya</i> (papaw)	<i>Persea americana</i> (avocado)
<i>Carthamus tinctorius</i> (safflower)	<i>Petroselinum</i> (parsley)
<i>Chenopodium</i> (Goosefoot)	<i>Phaseolus</i> (beans)
<i>Chenopodium quinoa</i> (quinoa)	<i>Pisum sativum</i> (pea)
<i>Chrysanthemum</i> (daisy)	<i>Poa</i> (meadow grass)
<i>Chrysanthemum indicum</i> (chrysanthemum)	<i>Prunus</i> (stone fruit)
<i>Cichorium intybus</i> (chicory)	<i>Prunus amygdalus</i>
<i>Citrullus lanatus</i> (watermelon)	<i>Prunus armeniaca</i> (apricot)

<i>Citrus</i>	<i>Prunus mume</i> (Japanese apricot tree)
<i>Colocasia esculenta</i> (taro)	<i>Prunus nana</i>
<i>Convallaria majalis</i> (lily of the valley)	<i>Prunus nigra</i> (Canada plumtree)
<i>Convolvulus</i> (morning glory)	<i>Prunus persica</i> (peach)
<i>Coriandrum sativum</i> (coriander)	<i>Prunus serotina</i> (black cherry)
<i>Crocus sativus</i> (saffron)	<i>Psidium guajava</i> (guava)
<i>Cucumis</i> (melons, cucumbers, gherkins)	<i>Punica granatum</i> (pomegranate)
<i>Cucurbita</i> (pumpkin)	<i>Raphanus sativus</i> (radish)
<i>Cucurbita moschata</i> (pumpkin)	<i>Rhus</i> (Sumach)
<i>Cucurbita pepo</i> (ornamental gourd)	<i>Rosa</i> (roses)
<i>Cuminum cyminum</i> (cumin)	<i>Rumex acetosa</i> var. <i>hortensis</i> (garden sorrel)
<i>Cydonia oblonga</i> (quince)	<i>Saccharum officinarum</i> (sugarcane)
<i>Cynara cardunculus</i> var. <i>scolymus</i> (globe artichoke)	<i>Secale cereale</i> (rye)
<i>Cyphomandra betacea</i> (tree tomato)	<i>Senecio</i> (Groundsel)
<i>Dahlia</i>	<i>Senecio vulgaris</i>
<i>Daucus carota</i> (carrot)	<i>Sesamum indicum</i> (sesame)
<i>Dianthus</i> (carnation)	<i>Solanum lycopersicum</i> (tomato)
<i>Dianthus caryophyllus</i> (carnation)	<i>Solanum melongena</i> (aubergine)
<i>Euphorbia</i> (spurges)	<i>Solanum nigrum</i> (black nightshade)
<i>Foeniculum vulgare</i> (fennel)	<i>Solanum tuberosum</i> (potato)
<i>Fragaria chiloensis</i> (Chilean strawberry)	<i>Spinacia oleracea</i> (spinach)
<i>Gladiolus hybrids</i> (sword lily)	<i>Trifolium</i> (clovers)
<i>Glycine max</i> (soyabean)	<i>Triticum</i> (wheat)
<i>Gossypium</i> (cotton)	<i>Tulipa</i> (tulip)
<i>Hemerocallis</i> (daylilies)	<i>Vicia</i> (vetch)
<i>Nasturtium officinale</i> (watercress)	<i>Vigna unguiculata</i> (cowpea)
<i>Nicotiana tabacum</i> (tobacco)	<i>Zea mays</i> (maize)
<i>Origanum majorana</i> (sweet marjoram)	

11.2 Appendix 2: Hosts of Green citrus aphid (*Aphis spiraecola*) as listed on the CAB Compendium (CAB International 2011)

Species	Species
<i>Abelia grandiflora</i> (Glossy abelia)	<i>Malus domestica</i> (apple)
<i>Annona</i>	<i>Nicotiana</i>
<i>Apium graveolens</i> (celery)	<i>Passiflora edulis</i> (passionfruit)
<i>Astragalus sinicus</i> (chinese clover)	<i>Persea americana</i> (avocado)
<i>Capsicum annuum</i> (bell pepper)	<i>Phaseolus vulgaris</i> (common bean)
<i>Carica papaya</i> (papaw)	<i>Pittosporum tobira</i> (Japanese pittosporum)
<i>Citrus</i>	<i>Prunus</i> (stone fruit)
<i>Citrus deliciosa</i> (Mediterranean mandarin)	<i>Prunus armeniaca</i> (apricot)
<i>Citrus limon</i> (lemon)	<i>Prunus persica</i> (peach)
<i>Citrus reticulata</i> (mandarin)	<i>Prunus salicina</i> (Japanese plum)
<i>Citrus sinensis</i> (navel orange)	<i>Pyrus communis</i> (European pear)
<i>Coriandrum sativum</i> (coriander)	<i>Saccharum officinarum</i> (sugarcane)
<i>Cotoneaster</i>	<i>Solanum melongena</i> (aubergine)
<i>Crataegus</i> (hawthorns)	<i>Solanum nigrum</i> (black nightshade)
<i>Cucumis sativus</i> (cucumber)	<i>Solanum tuberosum</i> (potato)
<i>Cucurbita</i> (pumpkin)	<i>Sorghum bicolor</i> (sorghum)
<i>Cydonia oblonga</i> (quince)	<i>Spiraea</i>
<i>Cynodon dactylon</i> (Bermuda grass)	<i>Spiraea thunbergii</i> (Thunberg's spiraea)
<i>Cyphomandra betacea</i> (tree tomato)	<i>Spiraea vanhouttei</i> (Bridal wreath)
<i>Daucus carota</i> (carrot)	<i>Theobroma cacao</i> (cocoa)
<i>Eriobotrya japonica</i> (loquat)	<i>Viburnum</i>
<i>Glycine max</i> (soyabean)	<i>Viburnum odoratissimum</i>
<i>Juglans regia</i> (walnut)	<i>Vigna unguiculata</i> (cowpea)
<i>Lactuca sativa</i> (lettuce)	<i>Vitis vinifera</i> (grapevine)
<i>Macadamia ternifolia</i> (Queensland nut)	<i>Zea mays</i> (maize)
<i>Malus</i> (ornamental species apple)	

11.3 Appendix 3: Hosts of Plum pox virus (*potyvirus*) as listed on the CAB Compendium (CAB International 2011)

Scientific name	Common name
<i>Cichorium</i> sp.	Chicory
<i>Cirsium arvense</i>	Creeping thistle
<i>Clematis</i> sp.	
<i>Convolvulus arvensis</i>	Bindweed
<i>Euonymus europaeus</i>	
<i>Juglans regia</i>	Carpathian walnut
<i>Ligustrum vulgare</i>	Privet
<i>Prunus americana</i>	American plum
<i>Prunus angustifolia</i>	Mountain cherry tree
<i>Prunus armeniaca</i>	Apricot
<i>Prunus avium</i>	Sweet cherry
<i>Prunus besseyi</i>	Bessey cherry
<i>Prunus cerasifera</i> *	Cherry plum, Myrobalan plum
<i>Prunus cerasus</i>	Sour cherry
<i>Prunus cistena</i>	
<i>Prunus davidiana</i>	
<i>Prunus domestica</i>	Plum
<i>Prunus dulcis</i>	Almond
<i>Prunus emarginata</i>	Bitter cherry tree
<i>Prunus fruticosa</i>	Dwarf cherry
<i>Prunus glandulosa</i> *	Almond cherry, Dwarf flowering almond
<i>Prunus hortulana</i>	
<i>Prunus humilis</i>	
<i>Prunus ilicifolia</i>	Holly-leaved cherry
<i>Prunus insititia</i> *	Damson plum
<i>Prunus japonica</i> *	Japanese single bush cherry
<i>Prunus maackii</i>	
<i>Prunus mahaleb</i> *	Mahaleb cherry, St. Lucie cherry
<i>Prunus maritima</i> *	Beach plum
<i>Prunus mexicana</i>	
<i>Prunus nigra</i>	Canada plumbtree
<i>Prunus padus</i>	Bird cherry
<i>Prunus pensylvanica</i>	Pin cherry

Scientific name	Common name
<i>Prunus persica</i>	Peach
<i>Prunus pumila</i> var. <i>besseyi</i>	
<i>Prunus pumila</i> var. <i>depressa</i>	
<i>Prunus salicina</i> *	Japanese plum
<i>Prunus sargentii</i>	Sargent's cherry
<i>Prunus serotina</i>	Black cherry
<i>Prunus serrulata</i>	Japanese flowering cherry
<i>Prunus sibirica</i> *	Siberian apricot
<i>Prunus spinosa</i> *	Blackthorn, Sloe
<i>Prunus subhirtella</i>	Weeping Japanese cherry
<i>Prunus tenella</i>	
<i>Prunus tomentosa</i> *	Nanking cherry
<i>Prunus triloba</i>	Rose tree of China
<i>Prunus virginiana</i>	Chokecherry tree
<i>Prunus virginiana</i> var. <i>demissa</i>	
<i>Prunus yedoensis</i>	
<i>Rorippa sylvestris</i>	Creeping yellowcress
<i>Solanum nigrum</i>	Black nightshade
<i>Sonchus</i> sp.	Sowthistle
<i>Taraxacum officinale</i> complex	Dandelion
<i>Trifolium</i> sp.	Clovers

An asterisk (*) indicates those species that are often grown in domestic gardens or used as street trees in Australia. These species would need to be considered in the event of an incursion of PPV (Diagnostic protocol for *Plum pox virus*).

11.4 Appendix 4: Standard diagnostic protocols

For a range of specifically designed procedures for the emergency response to a pest incursion refer to Plant Health Australia's PLANTPLAN (www.planthealthaustralia.com.au/plantplan).

11.5 Appendix 5: Resources and facilities

Table 11 provides a list of diagnostic facilities for use in professional diagnosis and advisory services in the case of an incursion.

Table 11. *Diagnostic service facilities in Australia*

Facility	State	Details
DPI Victoria – Knoxfield Centre	Vic	621 Burwood Highway Knoxfield VIC 3684 Ph: (03) 9210 9222; Fax: (03) 9800 3521
DPI Victoria – Horsham Centre	Vic	Natimuk Rd Horsham VIC 3400 Ph: (03) 5362 2111; Fax: (03) 5362 2187
DPI New South Wales – Elizabeth Macarthur Agricultural Institute	NSW	Woodbridge Road Menangle NSW 2568 PMB 8 Camden NSW 2570 Ph: (02) 4640 6327; Fax: (02) 4640 6428
DPI New South Wales – Tamworth Agricultural Institute	NSW	4 Marsden Park Road Calala NSW 2340 Ph: (02) 6763 1100; Fax: (02) 6763 1222
DPI New South Wales – Wagga Wagga Agricultural Institute	NSW	PMB Wagga Wagga NSW 2650 Ph: (02) 6938 1999; Fax: (02) 6938 1809
SARDI Plant Research Centre – Waite Main Building, Waite Research Precinct	SA	Hartley Grove Urrbrae SA 5064 Ph: (08) 8303 9400; Fax: (08) 8303 9403
Grow Help Australia	QLD	Entomology Building 80 Meiers Road Indooroopilly QLD 4068 Ph: (07) 3896 9668; Fax: (07) 3896 9446
Department of Agriculture and Food, Western Australia (AGWEST) Plant Laboratories	WA	3 Baron-Hay Court South Perth WA 6151 Ph: (08) 9368 3721; Fax: (08) 9474 2658

11.6 Appendix 6: Communications strategy

A general Communications Strategy is provided in Appendix 6 of PLANTPLAN (Plant Health Australia, 2010).

11.7 Appendix 7: Market access impacts

Within the AQIS PHYTO database (www.aqis.gov.au/phyto) export of some material may require an additional declaration regarding freedom from the virus. Should aphid transmitted viruses be detected or become established in Australia, some countries may require specific declaration. Latest information can be found within PHYTO, using an Advanced search “Search all text” for the particular virus.